

Proceedings of the Japanese Cancer Association
The 19th General Meeting
December, 1960

日 本 癌 学 会 記 事

第 19 回 総 会

昭和 35 年 12 月

THE JAPANESE CANCER ASSOCIATION

Nishi-Sugamo, Toshima-ku, Tokyo, Japan

March, 1961

THE JAPANESE CANCER ASSOCIATION

Officers (1960)

PRESIDENT: Yuzo Tazaki

EXECUTIVE COMMITTEE

Yuzo Tozaki (Treasurer)	Tomizo Yoshida (Editor)	Kunio Oota (Secretary)
Kaneyoshi Akaazaki	Tamaki Imai	Sanji Kishi
Masaru Kuru	Waro Nakahara	Katsuo Takeda
Nobujiro Takizawa	Hideo Yagi	

ADVISORY COMMITTEE

Shiro Akabori	Kaneyoshi Akazaki	Shigeyasu Amano
Chisato Araki	Takashi Fujii	Masao Fujimori
Tokuji Fujinami	Yukio Hamazaki	Susumu Hibino
Kazunari Higuchi	Tamaki Imai	Hajime Imanaga
Morizo Ishidate	Ryuzo Iwatsuru	Dennosuke Jinnai
Tamaki Kajitani	Juntaro Kamahara	Shigetsugu Katsura
Sanji Kishi	Toshio Kurokawa	Masaru Kuru
Takamitsu Kusunoki	Sajiro Makino	Kazumasa Masubuchi
Yasushi Mitani	Toru Miyaji	Kazuo Mori
Waro Nakahara	Komei Nakayama	Gen-ichi Ogawa
Hajime Okamoto	Kunio Oota	Hachiro Sato
Mitsuo Segi	Masami Suda	Kitasu Suzue
Katsuo Takeda	Nobujiro Takizawa	Hisashi Tauchi
Yuzo Tazaki	Masanobu Tomoda	Kempo Tsukamoto
Isamu Usubuchi	Takeo Wada	Hideo Yagi
Hisashi Yamaguchi	Hisao Yamashita	Tomizo Yoshida

OFFICE

THE JAPANESE CANCER ASSOCIATION

c/o Cancer Institute, Nishisugamo, Toshima-ku, Tokyo, Japan

CONTENTS

Proceedings of the XIXth General Meeting of the Japanese Cancer Association

I. Biochemistry

1. H. Miyazaki, K. Oho, H. Tsukasaki, S. Sashida, E. Yokoyama, A. Shinoda, J. Ikeda: Comparative study of constituting amino-acids of the nuclear protein in lung carcinoma and normal lung tissue by means of Stein Moore's column chromatography..... 1
2. A. Oikawa, T. Matsushima: Ethanolamine in normal liver, regenerating liver and hepatoma of rat. 2
3. M. Hatanaka, J. Takei, Y. Sugino, O. Hayashi: Deoxynucleosidic compounds in tumor cells. 2
4. Y. Sekine, A. Iijima, H. Hirai, K. Warabioka: Some chemical and immunochemical properties of the proteins of rat ascites hepatoma cells..... 3
5. T. Kawachi, Y. Okamura, T. Suzuki, N. Uesaki, S. Fujii, Y. Yamamura: Studies on tryptophan pynolase of tumor bearing animals. 5
6. S. Shiba, A. Terawaki, J. Dohi, I. Ito, M. Miyatake: Studies on the enzyme formation in tumor-bearing rats. 6
7. N. Sameshima, Y. Ohno, N. Nakamura, S. Kobayashi, T. Sato: On serum phosphohexose isomerase and phosphoglucomutase in cancer patients. 7
8. M. Inoue, A. Oikawa: Glycogen phosphorylase activities of tumors regenerating rat liver and suckling rat liver..... 8
9. H. Terayama, T. Yamada, M. Matsumoto: Behavior of the protein-bound dye, N-demethylase, catalase and xanthine oxidase activities of rat liver during the course of repeated administrations of 3'-Me-DAB. 8
10. H. Sawada, Y. Sugita, S. Teratani, M. Kato, I. Ishikawa, A. Izaki, K. Kamino, Y. Nakamura, T. Inoue, S. Ikeda, H. Sakamoto: Proteolytic enzyme in Ehrlich ascites carcinoma cell. 9
11. H. Sato, T. Taneda, H. Oyama: The influence of γ_2 -globulin on succinic acid dehydrogenase and nucleic acid metabolism of tumor bearing animal..... 10

12. H. Nakazato, K. Rokuo: The transmethylation reaction in some experimental tumors of rats.....	11
13. H. Sawada, Y. Sugita, S. Teratani, S. Ikeda, H. Sakamoto, G. Matsuo, M. Kato, I. Ishikawa, A. Izaki, K. Kamino, Y. Nakamura, T. Inoue: Studies on glucose metabolism of Ehrlich's mouse ascites carcinoma cells in ascitic plasm.	11
14. K. Yamagishi, E. Hiyama: On oxygen consumption of livers of tumor bearing animals.	12
15. T. Tomaru, M. Ohashi, F. Fukuoka: The effect of 4-nitroquinaldine N-oxide on DPN metabolism in tumor tissues.	12
16. H. Sato, K. Yunoki, S. Otsuji, T. Higashi, M. Moriyama, T. Ichiki, H. Oyama, T. Sameshima, S. Maeda, S. Maeda: Studies on the mechanism of haematoprotein metabolism in cancer (I).....	13
17. H. Sawada, Y. Sugita, G. Matsuo, S. Teratani, M. Katoo, I. Ishikawa, A. Izaki, K. Kamino, T. Inoue, Y. Nakamura, S. Ikeda, H. Sakamoto: Nucleotide in the liver of tumor host.	14
18. H. Sawada, Y. Sugita, S. Teratani, G. Matsuo, M. Kato, I. Ishikawa, A. Izaki, G. Kamino, Y. Nakamura, S. Ikeda, H. Sakamoto: Studies on the capillary permeability factor of Ehrlich carcinoma cells.	15
19. S. Tanaka, H. Shigekane, K. Hattori, Y. Kato, S. Suga: Fractionation of liver catalase depressing factor of rat ascites hepatoma 130 (Preliminary Report).....	15
20. H. Sato, K. Yunoki: Purification and partial characterization of cancer toxin.....	17
21. S. Goto, S. Inui: On the serial changes of ¹³¹ I-labeled serum protein incorporated to rats bearing DAB-induced hepatoma.....	18
22. H. Yamaguchi, S. Ishigami, E. Ikeda, T. Saegusa, Y. Kōno, T. Tsuzuki, B. Machii: Zinc metabolism in malignant tumor (I).....	18
23. S. Tokuoka, T. Fujiwara: Distribution of zinc in the various tissues of tumor bearing animals: (I) Studies with ⁶⁵ Zn.	20
24. A. Umayahara, T. Mizunoya, Y. Kuroda, Y. Yamamura: A study on the metabolism of zinc of Ehrlich ascites and solid tumor-bearing mice by means of radioactive zinc.....	21
25. B. Haga, T. Mizunoya, A. Umayahara, T. Kawachi, S. Fujii, Y. Yamamura: Studies on the up-take of ⁶⁵ Zn by the liver of tumor bearing animals and regenerating liver.	22
26. H. Yamaguchi, S. Ishigami, T. Kurahori, S. Nakano, T. Morii, S. Yamaguchi, Y. Watanabe: Iron metabolism in malignant tumor (IV)	23
27. K. Nakao, T. Maekawa, M. Hattori, T. Wada, T. Shirakura, S.	

- Matsumoto, N. Tashiro, T. Kamiyama: Clinical studies on the causes of anemia in cancerous diseases. (II)..... 24
28. M. Murakami, Y. Tomari, O. Ohara, Y. Yagi, I. Kawagishi: Inhibitory effect of plasma and ascitic fluid from Ehrlich ascites tumor Mouse upon cytotoxic activities of fatty acids. 25

II. Chemistry

29. M. Abe, Y. Anraku, Y. Masamune, D. Mizuno: An antitumor combination therapy combining substances inhibiting the synthesis of DNA, RNA and of protein..... 27
30. K. Hano, H. Iwata, A. Akashi: Influence of some carcinostatics on the activities of liver catalase, xanthine oxidase and uricase in normal and tumor bearing mice. (XXVII, XXVIII)..... 28
31. T. Ebina, K. Satoo, M. Watanabe, M. Sato, N. Okamura: On the respiration, glycolysis and incorporation of ^{32}P into RNA and DNA of Yoshida sarcoma cells cultivated *in vitro*. 29
32. S. Tokuoka, H. Morioka, S. Tanaka: The membrane potential of the human cancer and related cells (II). Effects of several antitumor substances on the membrane potentials of the MN. sarcoma cells. 30
33. S. Osato, H. Mori, M. Morita: The observation on the electron-microscopic pictures of cancer cells under the action of chemotherapeutic agents. 31
34. K. Horie, M. Makita, K. Sato: Histopathological studies on subcutaneously transplanted experimental osteogenic sarcoma..... 33
35. S. Shinosawa, H. Yasuda: Electronmicroscopic studies on Yoshida sarcoma and ascites hepatoma (AH 130 and 7974 strain): Effects of Nitromin (Nitrogen mustard N-oxide) 34
36. Y. Sakurai, H. Imamura, A. Moriwaki, H. Isaka, M. Ishidate: Studies on the drug resistance of tumors (I) Development of resistance to alkylating agents in ascites tumors. 35
37. Y. Sakurai, H. Satoh, H. Imamura, A. Moriwaki, H. Imai: Studies on the drug resistance of tumors. (II) Grade estimation of resistance..... 36
38. H. Isaka, H. Imamura, A. Moriwaki, Y. Sakurai: Studies on the drug resistance of tumors. (III) Population analysis of drug-resistant tumors. 37
39. M. Urabe, T. Mizukami, S. Tsunamura, S. Miyazaki, K. Watanabe, T. Tachibana: Studies on the chemotherapy of malignant tumors: Experimental studies using Yoshida sarcoma cells..... 38
40. I. Hirono: On the mechanism of resistance to Nitrogen mustard N-oxide

in ascites tumors.	39
41. J. Watanabe, K. Kajiware: Further observations on the development of resistance against anti-cancer agents: Establishment of Nitromin resistant ascites hepatoma (AH 130 R) and 6-Mercaptopurine resistant adenocarcinoma (A 755/6 MPR)	40
42. H. Tsukada, A. Inoue, M. Ezoe, S. Fujiwara, T. Onoe: Metabolism of cancer cells and its relation to anti-cancer chemicals (V) Reversibility of Nitromin resistancy and biochemical properties of the tumor cells.	41
43. A. Hoshino, S. Kurita, T. Okita, K. Kimura: Experimental studies on the transformation of resistance to Mitomycin C in Yoshida sarcoma (I) ...	42
44. H. Endo: Study on cross-resistance of Amethopterin.....	43
45. S. Inoki, T. Ono: Effect of anti-tumor and carcinogenic substances on drug-resistance transformation in <i>Trypanosoma gambiense</i>	44
46. T. Yoshida, M. Ishidate, Y. Sakurai, K. Sawatari, H. Imamura, H. Satoh, T. Tashiro: Experimental studies on chemotherapy of malignant growth employing Yoshida sarcoma animals. (XXII) Antitumor activity of derivatives of Nitrogen mustard containing only one chloroethyl group.	45
47. M. Ishidate, T. Yoshida, Y. Sakurai, I. Yamamoto, M. Aoshima, T. Tashiro, H. Satoh: Experimental studies on chemotherapy of malignant growth employing Yoshida sarcoma animals (XXIII). Antitumor action and toxicity in combination therapy of Nitromin and N,N'-dimethyl-N,N'-bis (2-chloroethyl) piperazinium dichloride.....	46
48. S. Hayashi, H. Ueki, M. Kaibara, M. Sakagawa: Studies on antitumor substances, on o-benzoquinone ethylenimine derivatives.	47
49. M. Maruyama: On the mode of anticancer action of RC4 (IV). Effect of several ethylenimines on oxidative phosphorylation by tumor mitochondria.	48
50. M. Arakawa, Y. Sato: Studies on the antitumor activity of seven-membered ring structures. (I)	48
51. K. Takano, Y. Hirokawa, Y. Kato, D. Mizuno, C. Ukita: Anticancer effects of Barbituric acid, Resorcinol, and Rhodan derivatives on ascitic and solid forms of Ehrlich tumor.	49
52. Y. Wada, N. Yamamoto, Y. Nakamura, S. Nakanishi, M. Ono: An experimental study on the antineoplastic action of d-glucosamine (I)	50
53. U. Matsuura: Effect of nucleic acid precursor on malignant neoplasm.....	51
54. H. Oshima: Studies on the anti-cancer activities of azo-compounds.	52
55. K. Sato, M. Sato, T. Nogi: Combination chemotherapy of cancer (V).....	53

56. S. Atsumi: Clinical studies on the anti-carcinomatous effect of epoxy compound unsaturated fatty acid.	54
57. S. Okada, Y. Uratsuji, M. Fukuda, K. Kimura, Y. Takahashi, Y. Tagashira, S. Hatano: Phase difference microscopic observation of the effects of various surfactants on the Ehrlich ascites carcinoma cells.....	54
58. H. Kinukawa, K. Tanikawa, J. Tanami, H. Hirano: The Oncolytic effect of the extract obtained from the stomach, intestine, mesentery and mesenterial lymphnodes.	55
59. N. Ishida, K. Kumagai, K. Miyazaki, M. Ito: Carzinostatin, a new anti-tumor substance.	56
60. T. Arai, H. Hori: Anti-tumor activity of three antibiotics derived from streptomyces.	57
61. S. Hosokawa, K. Yamashita, S. Watanabe, K. Nakagawa, S. Nakayama, T. Mishima, N. Kamano: Experimental studies of effects of synthetic steroids on cancer cells. (III) Anticancer Effects of stigmasterol....	58
62. K. Ueda: Anti-tumor action of earthed sera of tumor-bearing animals (XXX)	59
63. T. Kayama: Studies on an oncostatic agent "K.C.G." (I)	59
64. F. Shiomi, H. Onoda, K. Yamamoto: Experimental and clinical studies on the effect of K.C.G. (I) Effect of K.C.G. on normal rabbit & Yoshida sarcoma.	60
65. The speech was given up.	
66. The speech was given up.	
67. K. Nabeya, Y. Iijima: Clinical experience in use of anti-cancerous drug W.T.T.C.....	61
68. K. Hiraki, H. Sunami, K. Kitashima: Screening of various anti-leukemic agents by our bone-marrow tissue culture technic.....	62
69. J. Tokuyama, S. Mizota, H. Satou, T. Tsunematsu, H. Tokuyama, H. Satoh, T. Tashiro: Experimental and clinical studies on Nitromin D.....	63
70. T. Ebina, M. Sato, K. Sato, M. Watanabe, N. Okamura: Studies on the cytostatic activities of Endoxan, Trenimon, and E 39.	64
71. T. Mineshita, T. Iwaki, K. Yamaguchi, H. Tsujii: Antitumor effect of Endoxan on the transplantable and spontaneous animal tumors.....	65
72. J. Tokuoka, S. Mizota, H. Satou, T. Tsunematsu, H. Tokuyama, H. Satoh, T. Tashiro: Experimental and clinical studies on Endoxan.	66
73. Y. Tanakadate, I. Oohashi, H. Nakaseko: Clinical experience with Endoxan.	67
74. Y. Shiraha, K. Sakai, Y. Nakamura, K. Hashima: Clinical trials of	

Endoxan, a new anti-tumor agent.	67
75. Y. Takemasa, T. Sugiyama, T. Kimura, Y. Koyama: Studies on chemotherapy of malignant neoplasma (VIII).....	68
76. N. Iijima, K. Matsuura, A. Ueno, K. Fujita, T. Aiba, H. Ukishima, F. Nohara, T. Koshizuka: Fundamental and clinical studies on M.H.	69
77. H. Soeda, S. Okamura: Distribution of ^{203}Hg -hematoporphyrin- Na_2 in tumor bearing mice and co-action of X-ray and hematoporphyrin- Na_2	69
78. S. Tasaka, K. Mashimo, Y. Kuroda, T. Harada, K. Shimizu, M. Hatakeyama, O. Kunii, E. Yamada, T. Jindate, K. Shimada: Studies on Chromomycin, experimental and clinical and adenosine-deaminase activity of serum of cancer patient.	70
79. T. Hattori, G. Fujii, K. Ashikawa, K. Motoya, Y. Ishibashi: Chemotherapy in cancer patients treated with Mitomycin-C and bone marrow transplantation.	71
80. I. Usubuchi, S. Oboshi, J. Yoshida, M. Sugawara, T. Hongo: Studies on the intermittent application of Mitomycin C.	72
81. K. Okajima, N. Sakakibara, K. Morishita: Studies on the preoperative use of anti-carcinogenic agents (III): Adequate time for administration of anti-carcinogenic agents.....	74
82. K. Kawamura, S. Kitagawa, K. Nishimura, S. Kawamura, A. Oomoto, T. Sato, M. Nakajima, T. Ebise, R. Okamoto: Studies on local administration of anti-cancer agents by means of tissue culture and electronmicroscopy.	76
83. C. Ohta, M. Miura, Y. Harima, Y. Takeda: Experimental studies on local application of anticancerous chemotherapeutics in surgical operation (II)	77
84. R. Oono, M. Akagi, R. Tomojiri, H. Saitoo, K. Kitano, F. Katsuhisa: Experimental studies of anticancer agent combined with surgery.....	77
85. I. Ōshiro, S. Kusama, T. Yoshida, T. Iwasaki, K. Matsuoka: Chemotherapy of intracranial tumors.	78
86. T. Ohara: The study on the chemotherapy of gastric cancer: Preliminary report.	79
87. M. Ishihara, Y. Kawashima, T. Uchida, S. Ino, T. Nakatsuka, K. Kato, M. Miki: Chemical experiences of anticancer agents in gynaecological fields.	80
88. S. Yamamoto, Y. Kawashima, T. Uchida, T. Masukawa: On the five year survival rate in the cases of cancer of the uterine cervix which treated by 8-Azaguanine as additional treatment agent.	81

89. J. Oomura, K. Ookita, S. Tasaka: Clinical and pathological studies on cancer of urinary bladder with anticancer drugs..... 82
90. T. Kusumoto, J. Kobayashi, H. Kameyama: Study of the serum iron level for evaluation of antitumor agents..... 83
91. S. Inui, Y. Shimizu: Effects of anticancerous agents on the serum protein and conjugated protein fraction of patients with malignancy..... 84
92. M. Ogiwara, S. Yamada, S. Toshima, S. Shimizu, I. Kato, T. Umesono, F. Nakada, N. Tanaka: The method deciding the effect mechanism of anticancer agent by fluctuation of phosphatase value in ascites extracted from Yoshida sarcoma rats. 84
93. Y. Inazu, Y. Sugiya, M. Arakawa, S. Osamura: Studies on protection of leukopenia due to antitumour agents. (II): Effect of some leukocytosis-promoting substances on leukopenia due to RC-4. 86

III. Cytology

94. K. Morishita, K. Utsumi: A critic on NTS staining for cancer cells..... 87
95. T. Yoshida, H. Isaka, S. Odashima, M. Ishidate, T. Yamada: Studies on the ascites hepatoma. (XIX): Further studies on the chromosome..... 88
96. M. Takahashi, K. Hamada, H. Awaya, K. Inoue: Comparative idiogram analysis of MN-sarcoma cells (T-strain), maintained by continuous tissue culture and those maintained by serial animal passages. 89
97. H. Okumura, T. Takaoka, H. Katsuta: Studies on strain HeLa cells (cervical carcinoma of human uterus) in protein-free media. (II): Comparison of chromosomes among the cells cultivated in serum medium and two substrains in protein-free medium. 91
98. M. Kawana, K. Kawaguchi, T. Takaoka, M. Umeda, H. Katsuta: Studies on strain L cells (mouse fibroblasts) in protein-free media. (VIII): Effects of sera on the appearance of multinucleated cells..... 92
99. H. Okamura, T. Takaoka, H. Katsuta: Studies on strain L cells (mouse fibroblasts) in protein-free media. (IX): Comparison of chromosomes among four substrains cultivated continuously in protein-free media..... 92

IV. Histology

100. K. Okano, K. Nagai, H. Uda, Y. Mori, T. Saki, H. Taniguchi, G. Ueda, T. Fujimoto, F. Tarutani: Studies of inclusion bodies in human cancer cells. 94
101. Z. Ota, H. Matsunori: New intracytoplasmic and intranuclear cry-

stallizations in various tumor cells.	95
102. T. H. Yoshida: Invasive ability of diploid and tetraploid tumor cells in rats and mice.	96
103. T. Sato, H. Tauchi: On the age factor on the histological variation in the cancer tissues with special reference to the relationship between pictures of cancer cells and of interstitia.....	97
104. T. Nagayo, T. Komagoe: Histological studies on mucosal carcinoma of the stomach; Fundamental structures of the cancer tissues and their relationship to the histological pictures of the non-cancerized gastric mucosa.	98
105. M. Someya, K. Takagi: A clinico-pathological study of early ulcer- carcinoma.	99
106. H. Kitade, T. Morioka, T. Isohashi, K. Fukuda, K. Hayashi, S. Asada: Studies on gastric carcinoma with reference to the relationship between the histopathological nature of the carcinoma and the alteration in the cellular constituent of the regional lymph nodes.	101
107. T. Ogata, M. Kitamura: A histochemical study of the gastric cancer.	102
108. T. Ishidate: Pathologic-anatomical studies on carcinoma of the panc- reas with special reference to diabetes mellitus as a complication.....	103
109. T. Kato, J. Chidoi, T. Aoki: Study on the lymphatic follicle of the urological tumors.	104
110. M. Fujimori, M. Izuo: Histopathological relationships between can- cerous and mastopathic breasts in Japanese women.	104
111. F. Watanabe, I. Kuwatsuka, S. Ogata, M. Hashiguchi: 2 cases of spontaneous multiple mammary tumors with their metastasis in cats.....	106
112. N. Takizawa, B. Takeda: Studies on the atypical proliferation of cervical epithelium and cervical carcinoma.	107
113. T. Kawanishi: Histochemical study on carcinoma in situ of the uterine cervix.	108
114. S. Iwai, T. Saito, K. Shiozawa, T. Tsuda: Histological classification for staging of cervical cancer.	109
115. G. Izoe, J. Fujii: Histopathological study on carcinoma of the uterine body.	110
116. T. Kasuga, K. Oota: On the fine structures of adenoacanthoma of the endometrium.	111
117. N. Sasano, H. Wakasa: Histopathological studies on primary malignant tumors of the adrenal cortex and medulla.....	112
118. T. Miyaji, K. Ishida, N. Kambara, R. Tateishi, K. Watanabe, M. Sasaki, Y. Maruo, M. Miyamoto, S. Yui, S. Ishihara: Histological classifi-	

	cation of 414 cases of bronchogenic carcinoma.....	114
119.	K. Matsui, S. Moriwaki, T. Yamashiro: Electron-microscopic studies on maxillary carcinoma and normal and chronic inflammatory epithelium of the maxillary sinuses, especially on its squamous cell metaplasia.	116
120.	M. Seki, K. Terao: Electronmicroscopic studies on human cancer (I) Squamous cell carcinoma.....	116
121.	Y. Mori, M. Hashimoto, A. Komori, M. Kosaka, T. Shimoyama, K. Akashi: Electron-microscopic studies on chorionepithelioma.....	118
122.	T. Kusunoki, K. Kashiwai, H. Yano, T. Matsunaga: Electron microscopic studies on clear cell carcinoma of the kidney.....	119
123.	T. Teshima, K. Nakamura, T. Ishidate: Electron-microscopic observations on sarcoma induced in the subcutaneous tissue of the rat by administration of trypan blue.	120
124.	T. Chiba: On tumor of dogs.....	121
125.	A. Sugimoto: Histological and histochemical studies on the duct-ligated salivary glands in rats.	121

V. Transplantation

126.	S. Maeda: Metabolism of P in Ehrlich ascites tumor cells after destruction of the cells (I)	123
127.	M. Miyakawa, S. Iijima, H. Kishimoto, Y. Uno, H. Midorikawa: Experimental study of human cancer cell transplantation into the germ-free animals using millipore filter. (II).....	124
128.	Y. Yasumura, T. Kuwata: Celostamo de Musa Mama Tumoro (SATÔ-Kancero N-ro 1) en Kontinua Kulturo <i>in Vitro</i>	125
129.	H. Miyawaki, S. Ishii: Milky secretion in a transplanted mammary cancer of the dd Mouse. (I) Influence of sex hormones on the secretory activity and transplantability.	126
130.	H. Miyawaki, S. Ishii: Milky secretion in a transplanted mammary cancer of the dd Mouse. (II) Histochemical studies of lipids.	127
131.	J. Takeuchi: An effect of acid mucopolysaccharide on the transplantation and growth of tumor.	128
132.	R. Mizumoto, A. Noda: Antitumoural effect on transplantation of ascites hepatoma AH 130. (I)	129
133.	I. Nagai, W. Yoshida, Y. Tanaka, M. Tanaka: Survival, proliferation and morphology of NF sarcoma tissue exposed to liquid Nitrogen.....	130
134.	T. Kuwata, H. Murayama: Studies on ascitic conversion of fructose sarcoma in mice.	131

135. A. Shiraishi: Studies on the susceptibility of ddN mice to inoculation with C₅₈ leukemic ascites.132
136. K. Okada, N. Kawamura: Establishment of a new transplantable strain of murine lymphocytic leukemia (Ascites Form) OHO No. 1133
137. T. Kumamoto, S. Soejima: Further studies on the transplantation of a mixture of different strains of tumor cells.134

VI. Carcinogenesis

138. M. Urabe, T. Mizukami, S. Tsunamura, S. Miyazaki, K. Watanabe, T. Tachibana: Cancer and mesenchymal tissues (II).....135
139. S. Kano, H. Kobayashi: An autopsy case with many primary tumors.....136
140. K. Ogawa, A. Tsutsumi, K. Iwata: On the mechanism of the tumorigenization with feeding test of the Yoshida tumor.137
141. Y. Ii, T. Kato, Y. Morimura, K. Nasu, T. Yamauchi, T. Miyaji: 2 cases of cholangiocarcinoma induced by Thorotrast administered more than 20 years ago.....138
142. S. Irino, T. Kotsuka, M. Kibata: Studies on lymphatic tumor of RF mice induced by irradiation. (I).....139
143. K. Fukui, C. Nagata, A. Imamura, Y. Tagashira: On the relation between the electronic structure and carcinogenic and carcinostatic activities of Urethan and its related compounds.....139
144. S. Hakomori, H. Kawauchi, T. Ishimoda: Molecular abnormality of a certain γ -glycoprotein and their changes during cancer development141
145. M. Matsumoto, H. Terayama: Relation between the enzymatic N-demethylation and the carcinogenic activity of various aminoazo dyes. ...141
146. A. Hanaki, H. Terayama, S. Kanda, M. Ishidate: Separation and purification of the Polar dyes.142
147. T. Okamoto, M. Itoh: Reactivity of 4-nitrogroups in the reaction between 4-nitroquinoline N-oxide and related compounds.....143
148. K. Tokita, S. Nishimura, K. Seki: Experimental studies on disposition and carcinoma (I) Alteration of disposition and body reaction.144
149. K. Tokita, R. Ito, H. Kawamura, M. Yamada, Y. Miyazaki, H. Henomatsu, T. Nagaoka, S. Unemoto, C. Isono, K. Tosaka, K. Takashima, K. Nagata: Experimental studies on disposition and carcinoma (II) Body reactions in Yoshida sarcoma.145
150. K. Tokita, K. Tosaka, K. Nagata: Experimental studies on disposition and carcinoma. (III) Prevention of carcinogenesis by the alteration of disposition of the host.....146
151. M. Matsuyama, T. Nagayo: The effect of nicotinamide and diphos-

phopyridine nucleotide on azo dye and methylcholanthrene carcinogenesis.	147
152. S. Odashima: Development of liver cancers in the rat by 4-dimethylaminostilbene (DAS) feeding following initial 4-dimethylaminoazobenzene (DAB) feeding. (I) Examination of the rat fed initially with DAB for more than one month.	148
153. S. Odashima: On the histogenesis of the ear duct and liver cancers in the rat fed with 4-dimethylaminostilbene (DAS).	149
154. K. Asano: On enzymatic and cytological changes during the carcinogenesis of mice and rats (I)	150
155. K. Kawakatsu, M. Mori, Y. Okamoto, R. Oka: Histochemical studies during carcinogenesis in mouse skin induced by 20-methylcholanthrene (I) The localization of alkaline phosphatase, acid phosphatase, esterase and β -glycosidase during carcinogenesis.	151
156. K. Kawakatsu, M. Mori, Y. Okamoto, R. Oka: Histochemical studies during carcinogenesis in mouse skin induced by 20-methylcholanthrene (II) Histochemical demonstration of protein-bound sulfhydryl and disulfide groups and PAS reactions during carcinogenesis.	152
157. K. Kawakatsu, M. Mori, Y. Okamoto, R. Oka: Histochemical Studies during carcinogenesis in mouse skin induced by 20-methylcholanthrene (III) Histochemical demonstration of aminopeptidase during carcinogenesis.	153
158. T. Onoe, I. Suzuki, G. Takahashi, Y. Koseki, M. Noro: Changes in ultrastructures of rat liver cells during DAB carcinogenesis (II) Significance of characteristic changes in endoplasmic reticulum.	153
159. N. Ito, Y. Fukuoka, M. Marugami, H. Nakamura, H. Kitamura: On the relationship between congo red index and quantitative histology in the rat.	155
160. M. Muramatsu, T. Osuga, S. Takasu, Y. Araki, S. Tasaka: Studies on the pyruvate metabolism during experimental hepatocarcinogenesis (I) <i>In vivo</i> metabolism of pyruvate- 2^{14}C	156
161. T. Sato, Y. Tamaru, S. Kishi: Paper electrophoretic studies on asparaginase in the liver of rats fed 4-dimethylaminoazobenzene (DAB).	157
162. Y. Koseki, Y. Jinnohara, T. Hirota, A. Inoue, H. Honma: Metabolic pathology of rat liver during DAB carcinogenesis (II).	158
163. T. Mineshita, T. Iwaki, J. Maeda, K. Yamauchi, K. Nagai: Survey on the spontaneous tumor of inbred female mice of dd-s homogeneous strain.	159

164. K. Warabioka:	Framkallning av underhudssarkom hos råttor med upprepade insprutningar av vattenlösning av streptomycin- och dihydrostreptomycin sulfat komplex.	160
165. K. Higuchi, T. Hosokawa, M. Iwata, I. Shinozaki, T. Arihiro, M. Aoyagi:	Experimental carcinoma of the uterine cervix in C ₃ H mice by 3,4-benzpyrene.	161
166. T. Sato:	Ecological approaches to the cause of stomach and lung cancer....	162
167. M. Kanisawa, G. Ide:	The carcinogenic action of para-benzoquinone on the lung of mice by experimental inhalation (II).....	164
168. A. Yasuno:	Studies on the histogenesis of lung tumor induced by hydrazid.	165
169. H. Kitamura, H. Shindo:	The electronmicroscopical study of experimental pulmonary tumor in mice.	165
170. K. Takemoto:	Experimental studies of lung tumor in rat, given carcinogenic hydrocarbons in combination with several dust of materials.	166
171. N. Kambara, T. Miyaji, R. Tateishi, M. Sasaki, K. Watanabe, T. Yamauchi, G. Yanabu:	Primary site and invasion-form of lung cancer using large section specimens.	167
172. S. Oboshi:	The significance of scar in the development of peripheral pulmonary carcinoma.	169
173. N. Kambara, K. Nasu, S. Onishi, M. Sasaki, T. Miyaji:	A case of lung cancer associated with asbestosis.....	170

VII. Virus

174. T. Imai, H. Okano, Y. Nishiumi:	Electron-microscopic observation of chicken sarcoma caused by intrathymic inoculation of viruses.	171
175. K. Shimpo, E. Narimatsu, H. Higashi, K. Nishida, K. Muroya:	Experimental studies on histogenesis of viral tumor (III) Comparative studies on the three strains of chicken sarcoma.....	172
176. T. Takahashi, M. Ishizawa, T. Takahashi:	Chemical studies on Chiba strain chick sarcoma virus.	173
177. T. Kasuga, Y. Shirasu, H. Miyamoto:	Histological & electronmicroscopical studies of Friend disease.....	173
178. M. Ohashi:	The isolation of toxohormone from Friend virus infected mouse spleen.	175
179. Y. Tsubura, K. Ichiba, I. Kimura, K. Matsumoto:	Development of lymphatic tumor in ddO and dd ₁ mice produced by Moloney's leukemia virus, recovery of virus and some evidence of viral transmission	

to the progeny.	176
180. Y. Tsubura, H. Kitamura, K. Ichiba, I. Kimura, M. Marugami, K. Matsumoto, M. Nakazawa: Pathological study of Moloney's leukemia virus inoculated mice.	177
181. K. Matsui, S. Moriwaki, K. Watanabe: Electron-microscopic studies on mammary carcinoma of the C ₃ H, SL and ddN strains mice, with special reference to proliferation mode of virus particles.	178
182. Y. Hamazaki, M. Murakami: On the acceleration of breast cancer of C ₃ H mouse caused by the agent isolated from leukemic AKR mice.	179
183. The speech was given up.	
184. M. Miyazawa, A. Nishikawa, T. Ushio, S. Matsumura, K. Sugiyama, K. Ueno, T. Yamamoto, K. Nishioka, A. Oda, H. Kinugawa: Clinical studies employing the oncolytic virus "ED" concerning the therapy about hepatoma.	180

VIII. Diagnosis

185. T. Koosaki, N. Uezumi, S. Hasegawa, S. Nakagawa, Y. Kotani, K. Muraki: Studies on malignolipin.	182
186. S. Yamagata, I. Nakagawa: An evaluation for malignolipin test.	183
187. M. Mizokuchi, H. Kobayashi: The evaluation of detection of malignolipin in blood in the diagnosis of malignant tumor.	184
188. S. Hattori, M. Matsuda, K. Morimoto: Studies on fluorescent dye (NTS); Its affinity for malignant tumor cells and effects on the development of tumor.	184
189. S. Matsumoto: The principle of the Davis test.	185
190. N. Yamaguchi, T. Sasai, G. Wakisaka, M. Kakei: Polarographic protein wave of M ₁ -fraction of serum in malignancies.	186
191. K. Kubo, G. Wakisaka: Clinical study on serum mucoprotein in leukemic patient.	187
192. M. Matsubara, T. Ohshita, F. Mitsui, M. Harada: Collective examination for cancer.	188
193. H. Irie, K. Murakami, H. Soeda, K. Harada: Mass survey of upper G.I. tract with special consideration on the early diagnosis of gastric cancer.	189
194. K. Ariga, K. Takahashi, S. Tanaka, H. Sato: On the operated cases of gastric cancer by gastric mass screening.	190
195. K. Kubo, T. Sasai, G. Wakisaka, Y. Takagi: Study on the acetone supernatant fraction of cancerous gastric juice.	190
196. K. Iwasaki, S. Kobayashi, M. Akagi: Studies on the ninhydrin test	

of fractionated gastric juice.....	191
197. A. Atsumi, S. Mito: The cytologic diagnosis of cancer using gastric juice.	192
198. Y. Yoshida, R. Inoue: The trials in the cytological diagnosis of gastric cancer.	193
199. M. Miura, K. Ono, T. Sato, H. Nakayama, Y. Takeda, M. Kimura, M. Togashi, M. Kon, C. Ohta, S. Shibata, Y. Harima, Y. Narumi: A mass examination for detection of rectal cancer in Aomori Prefecture.....	194
200. Y. Tazaki, H. Tominaga: Clinical aspect on the growth of lung cancer....	194
201. Y. Tazaki, I. Furukawa, S. Miyajima, H. Furue, Y. Inada, H. Tominaga, J. Sato, M. Gomi, F. Tagaya: Clinical statistics on lung cancer.	195
202. A. Tabuchi, A. Fujiwara: Statistical observation of group examination for the uterine cancer done during past 6 years.	196
203. Y. Ashitaka, S. Isojima, A. Yagi, N. Amaki, T. Hasegawa: Activity of the organization for early diagnosis of the uterine cancer.	197
204. H. Fujimori, H. Kinoshita, S. Noda: Clinical results of colpomicroscope reformed and improved by us.	198

IX. Radiation

205. T. Kuwata, Y. Yasumura, M. Kanisawa: Studies on inactivation of fructose sarcoma cells by UV-irradiation.	199
206. M. Tsuchidate, T. Nagashima, T. Wagai, S. Hayashi: Studies on the destruction of ascites tumor cells by intense ultrasound.....	200
207. A. Inui, T. Tabata: A cytological observation on chromosome breaks in MN lymphosarcoma cells induced by X-irradiation.	201
208. M. Maeda, I. Nishibata: The cytological studies of MN-sarcoma. (II) Mitotic inhibitory effect of X-irradiation.....	202
209. S. Oboshi, T. Shinozaki: Experimental studies on acquired radio-resistance of tumor cells (sequel report).....	202
210. K. Hori, S. Mochizuki, Y. Fujino: Effects of α -irradiation upon enzyme systems of tumors.	203
211. S. Maeda: Biological effects of so-called irradiation toxic substance (C-X substance)	204
212. D. Jinnai, S. Tanaka, J. Shimizu, M. Ono, K. Okajima, J. Kobayashi, Y. Kuwahara: Clinical studies of "OX substance".	205
213. K. Hara, Y. Hamasaki, S. Sakai: X-ray therapy of carcinoma of the paranasal sinuses and nasal cavities.....	206
214. H. Yamashita, S. Amino, M. Gomi: Results of radiotherapy of cancer	

of the tongue.	207
215. H. Yamashita, H. Kobayashi: Radiation treatment of the cancer esophagus....	208
216. H. Irie, K. Onizuka, T. Yasuda, A. Kasahara: Radiation therapy of advanced stomach cancer.	211
217. K. Narabayashi, S. Kimura, K. Maeda, H. Maenishi, M. Manabe, S. Ito, Y. Izumi: Follow-up study on 582 cases of gastric cancer irradiated after surgical operation.....	211
218. O. Kojima, M. Watanabe: Study on the radiation therapy of carcinoma of the cervix uteri.	212
219. H. Fujimori, F. Yamada, S. Maeda, H. Kinoshita, M. Morimura: Clinical experiences of plastbalt-macro in cases of cancer of uterine cervix.....	213

X. Therapy

220. K. Nakayama, F. Yanagisawa, Y. Honda, T. Kamata, K. Suzuki, N. Takizawa, H. Kakei: Combined surgical radiative treatment for cancerous lesions with particular interest in prevention of recurrence. (Theoretical basis for the preoperative irradiation in the treatment of esophageal cancer).	214
221. T. Ito, F. Yanagisawa, Y. Honma, K. Suzuki, M. Iwatsuka, H. Suzuki, S. Tamama: Histopathological evaluation of cancer of the lower esophagus and cardiac end of the stomach.	215
222. T. Kajitani, T. Hoshino, K. Takagi, M. Nakamura: Cancer of the stomach -An analysis of 2343 cases-	217
223. M. Tomoda, M. Mizokuchi: The carcinoma development in the gastric stump after partial gastrectomy for benign gastroduodenal lesion.	218
224. T. Oiwa, T. Toda, H. Saito, Y. Haraguchi: The problem of pancreatic lymph nodes in gastric cancer surgery.....	220
225. Y. Hara, K. Katsuta, M. Takahashi, B. Ito, T. Hoshino: Clinical observation of 75 cases of primary cancer of the lung in our clinic in the last 5 years.....	220
226. R. Nagai, M. Sengoku, M. Yoshida, M. Ukai: The result of surgery of adrenals for advanced cancer of the breast.....	221
227. H. Yagi: Annual report of the Japanese Committee of Carcinoma of the Uterus. (II)	222
228. K. Masubuchi, H. Kubo, T. Suzuki: Carcinoma of the cervical stump.....	224
229. K. Akashi, M. Hashimoto, A. Komori, Y. Yamada: Studies on the vaginal systematic radical hysterectomy for cancer of the uterine cervix and the extraperitoneal procedure of lymphodectomy with	

	ligation of the pelvic vessels (II).	225
230.	K. Higuchi, T. Katoo, T. Tazawa, S. Ogawa, T. Kobayashi, K. Hasuda, S. Kobayashi, Y. Terashima, A. Tanaka, H. Nagumo: Clinical and pathological studies on the Krukenberg's tumor.	226
231.	G. Momose, S. Mitsuhashi: Follow up study on the tumor of the bladder.....	227
232.	T. Kusunoki, T. Sonoda, T. Ohkawa, M. Takeuchi: Clinical study of primary hyper-parathyroidism (parathyroid adenoma) in urology.....	228

XI. Growth

233.	T. Shimazu, T. Suematsu: Variation in enzymic activity with the mitotic rate.	230
234.	R. Takaki, Y. Sugi, K. Takano, K. Kaneko, M. Umeda, T. Takaoka, H. Katsuta, H. Endo, H. Enomoto: Studies on collagen formation in tissue culture. (III) On the collagen formation by the strain cells (JTC-4 and -6) originated in rats.	231
235.	M. Kuru, G. Koosaki, T. Matushita, E. Itoo, T. Ogawa, S. Nakamura: Further studies on "Oncotrephin", the mitosis promoting substance in malignant tissues.	232
236.	M. Kuru, G. Kōsaki, Y. Aoki, S. Morishita, H. Watanabe: Isolation of the mitosis promoting substance from regenerating rat liver and chick embryo.	233
237.	H. Katsuta, T. Takaoka, I. Tagaya: Tissue culture of monkey kidney cells. (I) On the nutritional requirements.	234
238.	T. Takaoka, H. Katsuta, K. Kaneko: Studies on strain L cells (mouse fibroblasts) in protein-free media. (VII) Amino acid requirements in synthetic media.....	235
239.	H. Endo, K. Wakabayashi, H. Enomoto, H. Katsuta, T. Takaoka: Studies on the action of hormones in tissue culture. (III) Leucine aminopeptidase activity of the strain HeLa (cervical carcinoma of human uterus) and the effects of sex hormones.	236
240.	S. Kozuka: Studies on the growth factors of HeLa cells by dialyzing culture methods.	237
241.	K. Takano, Y. Hirokawa: Cytopathogenic effect of antisera on mammalian cells.	238
242.	S. Kozuka, K. Kojima: Electronmicroscopic studies on the role of serum protein upon HeLa cells.....	239
243.	K. Kawai, Y. Kato: Injurious effects of visible ray irradiation on cultures of HeLa cells.....	240
244.	K. Okano, K. Nagai, Y. Mori: The effect of colchicine on the HeLa	

cell.	241
------------	-----

XII. Metastasis

245. J. Fujiwara: The motility of gastric cancer cells (continued)	242
246. I. Watanabe, K. Morikawa, T. Katsui, Y. Nagaoka, Y. Takayanagi, S. Yamakawa: Über das Stadium der Transplantation und Metas- tase von experimentellem Tumor von Pleura insbesondere die Im- plantation zur verschiedenen Organe.....	243
247. H. Sato, Y. (Kawashima) Hikichi, K. Hashimoto, T. Takahashi: Studies on metastasis of cancer (VIII) Experiments on lymph nodes metas- tasis.	243
248. T. Saito, H. Sato: Studies on metastasis of cancer (IX). Metastasi- zability of transplantable mammary carcinomas in C ₃ H mice.	244
249. T. Yamada: Intercellular adhesiveness of rat ascites hepatoma cells and metastasis. (II) Intercellular adhesiveness and negative electric surface charge in "islands" of ascites hepatomas during one transfer generation.....	245
250. K. Yoshida, H. Watanabe, K. Yamaguchi, A. Koizumi, S. Majima: An experimental study on metastasis formation to the liver.....	246
251. T. Yamada, L. Adachi, H. Isaka: The growth of different tumor strain in different organs following the transplantation into the left chamber of the heart.....	247
252. S. Kitamura, K. Kojima, K. Tsuchiya, S. Watanabe: Study on metas- tasis of gastric carcinoma. (A clinicopathologic classification of gastric carcinoma.).....	248
253. S. Takita, H. Nishijima, Y. Kishino, S. Toyota, T. Kawahigashi: A study of the mechanism of cancer metastasis.	250
254. K. Hayashi, S. Hirose, S. Ogawa, H. Sato: Experimental studies on the condition of the intramural communication of the tissue fluid in the gastro-duodenal wall.	251
255. H. Ikeuchi, S. Ogawa: Studies on the spread of the rectal cancer by means of resected specimens.....	252
256. T. Kobayashi, S. Takeuchi, K. Matsueda, K. Yamada: Patho-histolo- gical study on the relationship between primary lesions and lym- phnode metastasis in 163 cases of the operated patients with uterine cervical cancer.	253
257. H. Fujimori, Y. Kasahara: A statistical study on pelvic lymphnode metastasis of carcinoma of the cervix.....	254
258. N. Takizawa, K. Nakano: Studies on hematogenic metastasis of cancer....	255

259.	H. Munakata, T. Okawa, M. Osawa: Cytological criteria of cancer cells in the circulating blood.	256
260.	S. Majima, H. Watanabe, K. Yamaguchi, A. Koizumi, K. Yoshida: Clinical significance of tumor cells in the blood stream in patient with gastric carcinoma.	257
261.	M. Ohnishi, F. Yanagisawa, T. Kamata, I. Matsuzaki, M. Kawana, T. Yokoyama: Cancer cells in circulating blood of the patients with cancer of the esophagus (II)	257
262.	K. Wakasa, S. Kondo, K. Hashimoto: Cancer cells in the blood of lung cancer patients. (IV).....	258
263.	T. Kobayashi, S. Takeuchi, T. Kasamatsu, H. Koizumi, K. Matsueda, T. Yoshida, K. Yamada, M. Murakami, T. Sugimoto: Patients having gynecologic cancer with special reference to operative procedure.	259
264.	S. Hasegawa, Y. Wada: A study on the cancer cells in the peripheral blood of patients who received anticancerous drugs or radiation therapy....	260
265.	H. Saito, Y. Omori: Experimental study of the tumor cells in circulating blood.	261
266.	H. Satou, S. Mizota, H. Tokuyama: Effects of Nitromin on cancer cells circulating in the blood.	262

XIII. Immunity

267.	I. Usubuchi: Studies on the cross immunity among homologous tumor cells.	263
268.	I. Usubuchi, M. Sugawara, J. Yoshida: Studies on the cross immunity between heterologous tumor cells.	263
269.	S. Isojima: The effect of active immunization on development of mammary tumors in C ₃ H (Tax) mice (additional report)	264
270.	N. Takayanagi, Y. Miyake: Immunological analysis of the paraprotein fractions in the sera from patients with malignant tumors.	265
271.	M. Sekiguchi, K. Okada, M. Matsukura, Y. Ishibashi: Studies of the serum properdin levels in tumor-bearing patients and animals. (II).....	266
272.	K. Orita, K. Miyake: Studies on the relationship between serum properdin and cancer. (III) Effect of various drugs on the decrease of the serum properdin levels caused by antitumor agents.	268
273.	M. Aizawa: The effects of zymosan on human tumor growth in the conditioned rat.	269
274.	A. Tokunaga, S. Kijima, G. Kōsaki, J. Okamura: Mechanism of cytotoxicity of Ehrlich ascites tumour cells brought into contact with	

	the normal human serum (VII) The nature of the heat-labile factor.	270
275.	J. Okamura, H. Higashi, G. Kosaki: Mechanism of cytolysis of Ehrlich ascites tumour cells brought into contact with the normal human serum (VIII) The effect of natrium ions.	271
276.	K. Takeda, K. Kikuchi, K. Itakura, N. Tanigaki: Studies on the localization of the specific antigen in tumor cells.	272
277.	K. Takeda, Y. Tsuji, K. Maruyama, T. Maki: Immunological correlation between DAB hepatoma and MC sarcoma induced in an identical rat.....	273
278.	K. Takeda, Y. Fukushi, N. Tanigaki, K. Itakura: Studies on the tumor specific antigen using radioiodinated antiserum globulin.....	274
279.	K. Takeda, T. Kanemoto: Influences of additional transplantation or excision on growth of the Takeda sarcoma.	274

XIV. Case Report

280.	A. Kobayashi, S. Ōhashi, M. Kaneko: 3 cases of double cancers.	276
281.	N. Tanaka, W. Chen, N. Kuribayashi: An autopsy case of gastric carcinoma showing extremely severe generalized anasarca due to generalized embolic acroangitic carcinomatosis in the dermia.	277
282.	M. Kyogoku, T. Okukubo, S. Aoki: An autopsy case of carcinosarcoma which originated in the stomach.....	278
283.	H. Kobayashi, S. Kano: 2 autopsy cases of leiomyosarcoma derived from the gastrointestinal tract.	280
284.	M. Kaneko, S. Ōhashi, N. Fukushima: A biopsy case of vascular tumor of the posterior rectal wall resembling to bone giant cell tumor.	281
285.	N. Matsuoka, M. Sagara, Y. Hamada, M. Hamada: An autopsy case of malignant mesenchymoma of the omentum associated with dermatomyositis.....	282
286.	R. Sano, N. Hanada: A case of non-chromaffin paraganglioma in the retroperitoneum.	283
287.	N. Tanaka, W. Chen, N. Kuribayashi: Huge angiomyolipoma of the kidney, incidentally found at emergency operation: Report of 2 cases.	284
288.	Y. Kondo: A case of surgically removed osteogenic sarcoma of the urinary bladder.	286
289.	K. Suga, M. Hamano: An autopsy case of primary mast cell leukemia.....	286
290.	K. Terao: An autopsy case of erythroleukemia.	287
291.	K. Hizawa, N. Inui: An autopsy case of embryonal rhabdomyosarcoma	

in the left auricular region.	288
292. T. Okamoto, N. Kobayashi: An autopsy case of branchiogenic carcinoma.	289

SPECIAL LECTURE

I. Sarah E. Stewart: Polyoma virus as a model for studies on the viral etiology of human neoplasms.	291
II. Tadashige Murakami: On the point of the development of stomach cancer.....	305
III. Kunio Oota: On histogenesis of gastric carcinoma: A histopathological study on the genesis of scirrhous carcinoma of the stomach among the Japanese.	313

目 次

第19回 日本癌学会総会

I. 生 化 学

1. 宮崎秀樹, 於保健吉, 塚崎 鴻, 指田勢郎, 横山英三, 篠田 章, 池田二郎:
肺癌細胞核蛋白質および正常肺細胞核蛋白質の Stein Moore カラムクロマ
トグラフィによる Amino 酸の比較研究 1
2. 及川 淳, 松島泰次郎: ラットの肝癌および再生肝に含まれる Amine について ... 2
3. 畑中正一, 武居順子, 杉野幸夫, 早石 修: 腫瘍細胞における Deoxynucleoside
化合物 2
4. 関根幸哉, 飯島あき子, 平井秀松, 蔵岡小太郎: 腹水肝癌細胞構成蛋白質の化
学的免疫学的性質 3
5. 河内 卓, 岡村 裕, 鈴木友和, 上崎典雄, 藤井節郎, 山村雄一: 担癌動物の
肝臓 Tryptophan pyrrolase について 5
6. 芝 茂, 寺脇朝治, 土肥淳二, 伊藤一二, 宮武 実: 担癌動物における酵素
合成に関する研究 6
7. 鮫島夏樹, 大野義雄, 中村尚孝, 小林定造, 佐藤拓司: 癌患者の血清 Phos-
phohexose isomerase および Phosphoglucomutase について: 7
8. 井上正順, 及川 淳: 癌細胞および再生肝の Glycogen phosphorylase 活性
について: 8
9. 寺内 宏, 山田敏雄, 松本光男: 3'-Me-DAB 反復投与時のラット肝にみられ
る蛋白結合色素, Demethylase, Catalase および Xanthine oxidase 活
性の消長について 8
10. 沢田平十郎, 杉田幸男, 寺谷 進, 松尾源一郎, 加藤守彦, 石川育夫, 井崎
昭, 紙野建人, 中村義尚, 井上武彦, 池田周司, 坂本 弘: Ehrlich 腹水癌
細胞蛋白分解酵素 9
11. 佐藤八郎, 種子田哲郎, 大山治史: γ_2 -globulin の Ehrlich 腹水癌細胞および

	担癌マウス肝、腎の脱水素反応系および核酸代謝におよぼす影響 (第1報).....	10
12.	中里博昭, 六尾勝美: シロネズミの実験的腫瘍における Methyl 基転移反応.....	11
13.	沢田平十郎, 杉田幸男, 寺谷 進, 池田周司, 坂本 弘, 松尾源一郎, 加藤守彦, 石川育夫, 井崎 昭, 紙野建人, 中村義尚, 井上武彦: Ehrlich 腹水癌の腹水中における糖代謝	11
14.	山岸一一, 檜山栄一: 担癌動物肝臓の酸素消費について	12
15.	都丸禎三, 大橋望彦, 福岡文子: 腫瘍組織の DPN 代謝に対する 4-Nitro-quinaldine N-oxide の影響	12
16.	佐藤八郎, 柚木一雄, 尾辻義人, 東 達郎, 森山正武, 大山治史, 市来輝也, 鮫島哲也, 前田俊二, 前田鹿三: 癌における Haemo 蛋白体の代謝機構に関する研究 (第1報).....	13
17.	沢田平十郎, 杉田幸男, 松尾源一郎, 寺谷 進, 加藤守彦, 石川育夫, 紙野建人, 井上武彦, 中村義尚, 池田周司, 坂本 弘: Ehrlich 担癌マウスの肝核酸に関する研究	14
18.	沢田平十郎, 杉田幸男, 寺谷 進, 松尾源一郎, 加藤守彦, 石川育夫, 井崎 昭, 紙野建人, 中村義尚, 池田周司, 坂本 弘: Ehrlich 癌細胞の血管透過性因子に関する研究	15
19.	田中伸一, 茂兼英寿, 服部和彦, 加藤義昭, 須賀昭二: ラッテ腹水肝癌 130 の産生するマウス肝 Catalase 低下物質について (予報).....	15
20.	佐藤八郎, 柚木一雄: 癌腫毒の精製および化学的特性について	17
21.	後藤 俊, 乾 成美: 担癌シロネズミ体内における ¹³¹ I 標識血清蛋白の経時的变化について	18
22.	山口 寿, 石上重行, 池田悦郎, 三枝達明, 河野泰通, 都築敏男, 町井文吾: 郎: 悪性腫瘍の亜鉛代謝 (I).....	18
23.	徳岡俊次, 藤原 剛: 担癌体内の亜鉛分布に関する研究 (第1報) ⁶⁵ Zn による実験的研究	20
24.	馬屋原晟, 水納谷民太郎, 黒田吉男, 山村雄一: ⁶⁵ Zn による Ehrlich 腹水癌および固型癌の亜鉛代謝に関する研究	21
25.	芳賀 敏, 水納谷民太郎, 馬屋原晟, 河内 卓, 藤井節郎, 山村雄一: 担癌動物の肝臓および再生肝の ⁶⁵ Zn のとりこみについて	22
26.	山口 寿, 石上重行, 倉堀知弘, 中野俊一, 森井健, 山口清三, 渡辺善正: 悪	

性腫瘍の鉄代謝(第4報)	23
27. 中尾喜久, 前川 正, 服部理男, 和田武久, 白倉卓夫, 松本修一, 田代矩彦, 神山照秋: 悪性腫瘍の貧血に関する研究 (第2報)	24
28. 村上元孝, 泊 康男, 小原 修, 八木泰夫, 川岸一郎: 脂肪酸の細胞傷害能に 対する腹水癌マウスの血漿および腹水の阻害効果について	25

II. 化 学 療 法

29. 阿部美穂子, 安楽泰宏, 正宗行人, 水野伝一: DNA, RNA および蛋白質合 成の特異阻害剤の組合せによる抗癌性	27
30. 羽野 寿, 岩田平太郎, 明石 章: 正常および担癌動物の肝 Catalase, Xanthine oxidase ならびに Uricase 活性におよぼす諸種制癌薬の影響に ついて (第27, 28 報)	28
31. 海老名敏明, 佐藤和男, 渡辺民朗, 佐藤正弘, 岡村伸子: 培養吉田肉腫細胞の 呼吸, 解糖, 核酸代謝	29
32. 徳岡俊次, 森岡 久, 田中聖児: 超微小電極法による人腫瘍細胞膜電位に関す る研究(第2報)	30
33. 大里俊吾, 森 一, 森田倫雄: 種々なる制癌剤により微細構造のこうむる変 化の比較観察	31
34. 堀江健也, 牧田元雄, 佐藤計人: 継代皮下移植せる実験的骨原性肉腫の病理組 織学的研究	33
35. 篠沢貞夫, 安田寛基: 吉田肉腫および腹水肝癌(AH 130, 7974) の電子顕微 鏡的研究, とくに抗癌剤(Nitromin) に対する影響について	34
36. 桜井欽夫, 今村 博, 森脇絢子, 井坂英彦, 石館 基: 腫瘍の薬剤耐性の研究 (第1報) 腹水腫瘍の Alkyl 化剤に対する耐性の発現	35
37. 桜井欽夫, 佐藤 博, 今村 博, 森脇絢子, 今井弘子: 腫瘍の薬剤耐性の研究 (第2報) 耐性度の測定	36
38. 井坂英彦, 今村 博, 森脇絢子, 桜井欽夫: 腫瘍の薬剤耐性の研究 (第3報) 耐性腫瘍の Population Analysis	37
39. 卜部美代志, 水上哲次, 綱村史郎, 宮崎誠示, 渡辺恵市, 橘 貞亮: 悪性腫瘍 の化学療法に関する研究, とくに Nitromin 抵抗性吉田肉腫を使用せる実 験的研究	38

40.	広野 巖: Nitromin に対する各系腹水腫瘍およびその耐性株の抵抗性の機序について	39
41.	渡辺順明, 梶原 強: 制癌剤に対する耐性発現に関する所見補足 (腹水肝癌 AH-130 の Nitromin 耐性株および Adenocarcinoma 755 の 6-MP 耐性株について)	40
42.	塚田英之, 井上愛子, 江副みよ, 藤原誠喜, 小野江為則: 癌の代謝と制癌剤の作用機構 (第5報) Ehrlich 腹水癌の Nitromin 耐性の可逆性と生物学的生化学的性状の変化	41
43.	星野 章, 栗田宗次, 大北 威, 木村喜代次: 吉田肉腫の Mitomycin C 耐性への型質転換に関する実験的研究 (第1報)	42
44.	遠藤英夫: Amethopterin の交叉耐性に関する研究	43
45.	猪木正三, 小野忠相: Trypanosoma 原虫の薬剤耐性 Transformation におよぼす抗腫瘍性物質および発癌性物質の効果	44
46.	吉田富三, 石館守三, 桜井欽夫, 猿渡健市, 今村 博, 佐藤 博, 田代田鶴子: 吉田肉腫を用いた悪性腫瘍の化学療法に関する実験的研究 (第22報) Mono-2-Chloroethylamine 誘導体の抗腫瘍効果	45
47.	石館守三, 吉田富三, 桜井欽夫, 山本 巖, 青島迪子, 田代田鶴子, 佐藤 博: 吉田肉腫を用いた悪性腫瘍の化学療法に関する実験的研究 (第23報) Nitromin および N,N'-dimethyl-N,N'-bis (2-chloroethyl) piperazinium dichloride の混合投与の制癌効果およびその毒性について	46
48.	林清五郎, 植木 寛, 甲斐原守夫, 坂川正之: 制癌物質の研究, ことに o-Benzoquinone ethylenimine 誘導体について	47
49.	丸山素弘: RC 4 の制癌機構の研究 (第4報) 癌細胞の酸化的燐酸化におよぼす Ethylenimine 系 Alkyl 化剤の影響	48
50.	荒川順生, 佐藤裕信: 七員環化合物の制癌作用 (第1報)	48
51.	高野宏一, 広川康子, 加藤好雄, 水野伝一, 浮田忠之進: Barbituric acid, Resorcinol, Rhodan 誘導体の制癌効果	49
52.	和田義夫, 山本直明, 中村有行, 中西純夫, 小野政道: Glucosamine の抗腫瘍性についての実験的研究 (第1報)	50
53.	松浦梅春: 悪性腫瘍におよぼす核酸前駆物質の影響	51
54.	大島秀彦: Azo 系物質の制癌効果について	52

55.	佐藤 宏, 佐藤宮彦, 野木東洋: 抗癌物質併用療法の研究 (第 5 報).....	53
56.	渥美 理: Epoxy 結合不飽和脂肪酸製剤の抗腫瘍作用に関する臨床的研究.....	54
57.	岡田 聡, 裏辻嘉行, 福田素夫, 木村 健, 高橋洋介, 田頭幸夫, 波多野輔 久: 10 余種の界面活性剤処置による Ehrlich 腹水癌細胞の変性についての 位相差顕微鏡学的観察	54
58.	衣川淵水, 谷川久治, 田波潤一郎, 平野英夫: 胃腸管系および腸間膜, 腸間膜 淋巴腺抽出物質の制癌効果に関する研究	55
59.	石田名香雄, 熊谷勝男, 宮崎圭三, 伊藤政志: 新抗癌性物質 "Carzinostatin".....	56
60.	新井 正, 堀 宏行: 放線菌より得られた 3 種の制癌物質とその抗腫瘍作用.....	57
61.	細川修治, 山下貢司, 渡辺貞雄, 中川桂子, 中山 純, 三嶋登志夫, 釜野徳 明: 制癌に関する実験的研究. 特に合成ステロイドホルモンの制癌に関する 研究 (第 3 報) 大豆 Sterin の制癌効果	58
62.	上田幸蔵: 末梢血液の赤土処理における抗癌作用の検索 (第 30 報).....	59
63.	香山時彦: 抗腫瘍性物質 K.C.G. について (第 1 報)	59
64.	汐見文隆, 鉄田 寛, 山本 清: K.C.G. の実験と臨床 (第 1 報) K.C.G. の 正常家兎に対する作用および吉田肉腫に対する効果について	60
65.	都合により中止	
66.	都合により中止	
67.	鍋谷欣市, 飯島嘉之: 抗癌剤 W.T.T.C. の臨床経験.....	61
68.	平木 潔, 角南 宏, 喜多島康一: 教室考案の臨床組織培養法による抗白血病 剤のスクリーニング・テストについて	62
69.	徳岡淳一, 溝田 成, 佐藤 博, 常松 匠, 徳山英太郎, 佐藤 博, 田代田鶴 子: Nitromin D に関する実験的臨床的研究	63
70.	海老名敏明, 佐藤正弘, 佐藤和男, 渡辺民朗, 岡村伸子: Endoxan, Treni- mon, ならびに E 39 の制癌作用に関する基礎的研究	64
71.	峰下鏡雄, 岩城 徹, 山口健二, 辻井平三: Endoxan の動物腫瘍に対する作用.....	65
72.	徳岡淳一, 溝田 成, 佐藤 博, 常松匠, 徳山英太郎, 佐藤 博, 田代田鶴 子: Endoxan に関する実験的, 臨床的研究	66
73.	田中館義良, 大橋伊佐次, 中迫 博: Endoxan の臨床効果	67
74.	白羽弥右衛門, 中村義扶, 酒井克治, 橋間健一: 抗癌剤 Endoxan の臨床応用.....	67
75.	武正勇造, 杉山 正, 木村 正, 小山善之: 悪性腫瘍の治療に関する研究	

(第8報)	68
76. 飯島 登, 松浦 潔, 上野 明, 藤田吉四郎, 相羽達雄, 浮島仁也, 野原不二夫, 塚塚為久: M.H. の基礎ならびに臨床的研究	69
77. 添田博彬, 岡村重昭: 担癌マウスにおける ^{203}Hg -Hematoporphyrin- Na_2 の体内分布および線と Hematoporphyrin- Na_2 との協同作用	69
78. 田坂定孝, 真下啓明, 黒田善雄, 原田敏雄, 清水喜八郎, 畠山正己, 国井乙彦, 山田栄八郎, 陣立恒夫, 島田 馨: Chromomycin に関する実験的ならびに臨床的研究および癌患者血清 Adenosine-deaminase 活性について	70
79. 服部孝雄, 藤井源七郎, 芦川和高, 元谷喜久雄, 石橋幸雄: 骨髓移植を併用した癌の Mitomycin 療法	71
80. 白淵勇, 大星章一, 吉田順之助, 菅原道義, 本郷敏郎: Mitomycin C の間歇投与に関する研究	72
81. 岡島邦雄, 榊原 宣, 森下和郎: 制癌剤の術前使用に関する研究 (第3報) 制癌剤の投与時期について	74
82. 河村謙二, 北川司良, 西村耕治, 河村章治, 大本 昭, 佐藤 透, 中嶋正温, 海老瀬尚文, 岡本良平: 制癌剤局所応用時の組織培養ならびに電子顕微鏡的研究	76
83. 太田忠造, 三浦光恵, 張間行直, 武田由紀夫: 制癌剤の手術野投与に関する検討 (第2報) 制癌剤, 抗菌剤併用の細菌増殖におよぼす影響	77
84. 大野隆二, 赤木正信, 友尻諒弥, 斎藤 寿, 勝久文雄, 北野邦俊: 制癌剤の外科手術併用に関する基礎的研究	77
85. 大城 勲, 草間 悟, 吉田 耕, 岩崎 隆, 松岡健司: 脳腫瘍の化学療法	78
86. 小原辰三: 胃癌の化学療法に関する研究 (第1報)	79
87. 石原 実, 川島吉良, 内田 正, 井篁重彦, 中塚 勉, 加藤和子, 三木 充: 婦人科領域における諸制癌剤の使用経験	80
88. 山元清一, 川島吉良, 内田 正, 益川照夫: 化学療法 (8-Azaguanine) 施行子宮頸癌患者の5年治療率について	81
89. 大村順一, 大北健逸, 田坂純雄: 制癌剤による膀胱腫瘍像の臨床病理学的変貌について	82
90. 楠本 剛, 小林淳一, 亀山英之: 制癌剤の効果判定の指標としての血清鉄について	83

91. 清水康世, 乾 成美: 悪性腫瘍患者の血清蛋白ならびに複合蛋白におよぼす抗癌剤の影響	84
92. 荻原正雄, 山田 欽, 戸嶋しまえ, 清水 進, 加藤 勲, 梅園 忠, 中田藤重, 田中信夫: 吉田肉腫ラッテ腹水の Phosphatase 値の変動を用いての抗癌剤の治療効果判定の一方法	84
93. 稲津佳彦, 杉谷幸男, 荒川順生, 長村重之: 抗癌剤使用時における白血球減少防止対策の研究 (第2報) RC-4 と白血球増多促進物質併用による効果について(2)	85

III. 細 胞

94. 森下和郎, 内海耕髓: NTS 染色の吟味	87
95. 吉田富三, 井坂英彦, 小田島成和, 石館 基, 山田 喬: 腹水肝癌の研究 (第19報) 各系の染色体について	88
96. 高橋 栄, 浜田克巳, 栗屋博信, 井上一男: <i>In vivo</i> および <i>in vitro</i> で継代した MN 系肉腫 (T 株) における核型の比較	89
97. 奥村秀夫, 高岡聡子, 勝田 甫: 無蛋白培地による HeLa 株細胞 (人子宮頸癌) の研究 (第2報) 血清培地継代細胞と無蛋白培地継代2亜株間の染色体の比較	91
98. 川名 衛, 川口一江, 高岡聡子, 梅田 誠, 勝田 甫: 無蛋白培地による L 株細胞 (マウス線維芽細胞) の研究 (第8報) 多核細胞形成に対する血清の影響	92
99. 奥村秀夫, 高岡聡子, 勝田 甫: 無蛋白培地による L 株細胞 (マウス線維芽細胞) の研究 (第9報) 無蛋白培地内継代4亜株間の染色体の比較	92

IV. 組 織

100. 岡野錦弥, 永井清和, 宇多弘次, 森 陽一, 佐木武夫, 谷口春生, 上田外幸, 藤本常彦, 樽谷文彦: 人体癌細胞封入体の研究	94
101. 太田善介, 松森 宏: 腫瘍細胞内に生ずる新結晶体について	95
102. 吉田俊秀: 二倍性および四倍性腫瘍細胞における組織浸潤性の差異	96
103. 佐藤秩子, 田内 久: 癌組織像の形態変換における年令的要約について (とくに癌細胞と間質との形態学的関連性を中心にして)	97

104.	長与健夫, 駒越喬貞: 胃粘膜癌の組織学的研究, 癌組織の基本像と非癌胃粘膜像との関係について	98
105.	染谷 守, 高木国夫: 早期胃潰瘍癌の臨床病理学的研究	99
106.	北出文男, 森岡哲吾, 磯橋 保, 福田勝次, 林 雄俊, 麻田 栄: 胃癌の浸潤形態と所属淋巴節内細胞の変動との関係について	101
107.	緒方卓郎, 北村元男: 胃癌の組織化学的研究 (癌細胞の Succinoxidase, DPN-diaphorase, TPN-diaphorase, α -Glycerophosphate dehydrogenase, Phosphorylase の活性について)	102
108.	石館卓三: 膀胱癌の病理解剖学的研究, とくに併発糖尿病との関連について	103
109.	地土井襄瑩, 加藤篤二, 青木 忠: 泌尿器腫瘍組織中の淋巴汭胞について	104
110.	藤森正雄, 泉雄 勝: 乳癌と乳腺症の組織学的関連についての再検討	104
111.	渡辺文友, 歟塚郁美, 尾形 親, 橋口ミドリ: 猫の多発性乳腺腫瘍の2例とその転移について	106
112.	滝沢延次郎, 武田 敏: 子宮腔部異型上皮と癌腫の研究	107
113.	河西孝信: 子宮頸部における上皮内癌の組織化学的研究	108
114.	岩井正二, 斎藤長士, 塩沢久要, 津田達雄: 子宮頸癌進行度の組織学的分類	109
115.	井添五郎, 藤井純一: 子宮体部癌に関する病理組織学的研究	110
116.	春日 孟, 太田邦夫: ヒトの子宮内膜腺棘細胞癌の微細構造について	111
117.	笹野伸昭, 若狭治毅: 副腎皮質および髄質の原発性腫瘍について	112
118.	宮地 徹, 石田健蔵, 神原信明, 建石竜平, 渡辺幸司, 佐々木基蔵, 丸尾泰生, 宮本 誠, 唯 正一, 石浜信太郎: 1958-1959 年 92 年間に剖検された肺癌 414 例の組織学的分類について	114
119.	松井敬介, 森脇昭介, 山城貞治: 上顎癌ならびに正常慢性炎症性上顎洞粘膜に関する電子顕微鏡的研究, とくにその扁平上皮化生について	116
120.	関 正利, 寺尾 清: 人癌の電子顕微鏡的研究(第1報) 角化扁平上皮癌について	116
121.	森 和郷, 橋本正淑, 小森 昭, 香坂三男, 下山利雄, 明石勝英: 絨毛上皮腫の電子顕微鏡的研究	118
122.	楠 隆光, 柏井浩三, 矢野久雄, 松永武三: 腎癌の電子顕微鏡的研究	119
123.	手島貞一, 中村克宏, 石館卓三: Trypan 青投与により発生したラッテ皮下肉腫の電子顕微鏡的研究	120

124.	千葉胤孝：犬の腫瘍について	121
125.	杉本顕俊：結紮唾液腺における再生増殖	121

V. 移 植

126.	前田茂和： ^{32}P 標識腹水癌細胞移植後の ^{32}P の運命 (第1報)	123
127.	宮川正澄, 飯島宗一, 岸本英正, 宇野 裕, 翠川 宏：ミリボールフィルター を利用して人癌細胞の無菌モルモットへの移植実験	124
128.	安村美博, 桑田次男：マウス乳癌(佐藤癌1号)細胞の組織培養による継代	125
129.	宮脇英夫, 石井昌三：マウス可移植性乳癌の乳汁様分泌について (第1報) 分 泌ならびに移植の成立におよぼす性 Hormone の影響	126
130.	宮脇英夫, 石井昌三：マウス可移植性乳癌の乳汁様分泌について (第2報) 脂 肪の組織化学的検索により見出される分泌乳汁の特殊性について	127
131.	竹内 純：腫瘍の移植および増殖に対する酸性多糖類の影響について	128
132.	水本竜二, 野田 彰：移植腫瘍における抗腫瘍性について (第1報)	129
133.	永井巖, 吉岡 済, 田中良憲, 久米川正好：凍結腫瘍組織の移植に関する実験 的研究	130
134.	桑田次男, 村山 浩：果糖肉腫の腹水型転換に関する研究	131
135.	白石彰徳： C_{58} マウスの腹水型リン巴球性白血肉腫の ddN マウスへの移植につ いて	132
136.	岡田耕一, 川村範夫：可移植性マウスリン巴球性白血病(腹水型)の一新系 OHO No. 1 の樹立	133
137.	熊本 亨, 副島清治：異った種類の腫瘍細胞の混合移植に関する研究 (続報)	134

VI. 発 癌

138.	卜部美代志, 水上哲次, 綱村史郎, 宮崎誠示, 渡辺恵市, 橋 貞亮：癌と間葉 組織系 (第2報)	135
139.	加野 敏, 小林久人：一個体に多発した原発性腫瘍の一剖検例	136
140.	小川勝土, 堤 啓, 岩田克美：吉田腫瘍試食による発癌機構について	137
141.	井 洋平, 加藤次男, 森村義行, 那須健治, 山田孝行, 宮地 徹：約 20 数年 前に Thorotrast を注射した患者に発生して胆管癌の2症例	138
142.	入野昭三, 小塚 堯, 木畑正義：X 線照射により惹起せしめた RF 系マウス	

の淋巴性腫瘍に関する研究 (第1報).....	139
143. 福井謙一, 永田親義, 今村 詮, 田頭勇作: Urethan およびその関連化合物 の電子状態と発癌および制癌作用について	139
144. 箱守仙一郎, 川内宏明, 石母田泰子: γ -異常糖蛋白体とその癌発育中変化	141
145. 松本光男, 寺山 宏: 各種 Aminoazo 色素の酵素的 N-demethyl 化と発癌 性の関係	141
146. 花木 昭, 寺山 宏, 神田昌一, 石館守三: Polar-dye (極性色素) の分離お よび精製	142
147. 伊藤道也, 岡本敏彦: 4-Nitroquinoline N-oxide 類における 4-位 nitro 基 の SH 化合物との反応性.....	143
148. 戸木田菊次, 西牟田祐昭, 関 一子: 癌と素質に関する実験的研究 (第1報) 素質の変革と生体反応	144
149. 戸木田菊次, 伊藤隆太, 川村弘徳, 山田光雄, 宮崎祥子, 舩松 洋, 長岡敏 雄, 畠本昌介, 磯野千冬, 登坂邦雄, 高島啓昌, 長田漢洵: 癌と素質に関す る実験的研究 (第2報) 吉田肉腫移植ラットの生体反応.....	145
150. 戸木田菊次, 登坂邦雄, 長田漢洵: 癌と素質に関する実験的研究 (第3報) 宿主の素質変革による癌発生の予防	146
151. 松山睦司, 長与健夫: Nicotinamide ならびに Diphosphopyridine nucleotide の Azo 肝癌および Methylcholanthrene 肉腫発生におよぼす影響	147
152. 小田島成和: 4-Dimethylaminoazobenzene (DAB) 短期間飼与後, 4-Dimethyl- aminostilbene (DAS) 飼与によるシロネズミ肝癌の発生について (第1報) DAB を1カ月以上与えた場合	148
153. 小田島成和: 4-Dimethylaminostilbene (DAS) 飼与によるシロネズミ外耳道 癌および肝癌の発生について	149
154. 浅野健夫: 実験的発癌過程における酵素学的細胞学的変化について	150
155. 川勝賢作, 森 昌彦, 岡本吉弘, 岡 隆一: マウス皮膚における実験的発癌過 程の組織化学的研究 (第1報) Alkaliphosphatase, Esterase ならびに β -Glycosidase について	151
156. 川勝賢作, 森 昌彦, 岡本吉弘, 岡 隆一: マウス皮膚における実験的発癌過 程の組織化学的研究 (第2報) SH, SS および PAS 反応について	152
157. 川勝賢作, 森 昌彦, 岡本吉弘, 岡 隆一: マウス皮膚における実験的発癌過	

程の組織化学的研究 (第3報) Aminopeptidase について	153
158. 小野江為則, 鈴木伊佐夫, 高橋五平, 小関弥平, 野呂昌熙: DAB 肝癌発生過程における肝細胞微細構造の変化 (第2報) 小胞体の特異的变化の意義	153
159. 伊東信行, 福岡善晃, 丸上昌男, 中村 博, 北村 旦: Azo 色素発癌過程におけるダイコクネズミ肝組織の量的変化と Congo red 係数との相互関係について	155
160. 村松正実, 大菅俊明, 多賀須幸男, 荒木嘉隆, 田坂定孝: 実験的肝癌発生過程における Pyruvin 酸代謝に関する研究 (第1報) Pyruvate-2-C ¹⁴ の In vivo の代謝	156
161. 佐藤永雄, 田村 裕, 岸 三二: Butter yellow 投与ネズミ肝 Asparaginase の濾紙電気泳動的研究	157
162. 小関弥平, 陣野原庸一, 広田映五, 井上愛子, 本間 宏: DAB 肝癌発生過程におけるラッテ肝の代謝病理 (第2報)	158
163. 峰下鏡雄, 岩城 徹, 前田甚作, 山口健二, 永井清和: dd-s 均一系マウスの自然発生腫瘍について	159
164. 蔵岡小太郎: 複合 Streptomycin (硫酸 Streptomycin および硫酸 Dihydrostreptomycin) 水溶液の反復皮下注射によるラッテ皮下肉腫の発生について ...	160
165. 樋口一成, 細川 勉, 岩田正晴, 篠崎一雄, 有広忠雅, 青柳昌樹: 3,4-Benzopyrene による C ₃ H マウスにおける子宮頸部の発癌実験	161
166. 佐藤徳郎: 胃および肺癌の成因に対する生態学的アプローチ	162
167. 蟹沢成好, 井出源四郎: Para-Benzoquinone 長時間連続吸入による実験的肺癌の発生について	164
168. 康野 明: Hydrazid 肺腫瘍の組織発生について	165
169. 北村 旦, 進藤裕之: 実験的マウス肺腫瘍の電子顕微鏡的研究 (第1報) 腫瘍発生前における肺胞壁の細胞反応	165
170. 竹本和夫: 癌原性炭化水素と各種粉塵との合併投与によるラット肺腫瘍の実験的研究	166
171. 神原信明, 宮地 徹, 建石竜平, 佐々木基義, 渡辺幸司, 山内孝行, 柳父暁二: 原発性肺癌の発生部位および肺内進展について	167
172. 大星章一: 肺周辺部癌の発生における癒痕の意義について	169
173. 神原信明, 那須健治, 大西俊造, 佐々木基義, 宮地 徹: 石綿肺に合併せる肺	

癌の一例	170
------------	-----

VII. Virus

174. 今井 環, 岡野博光, 西海雄幸: 家鶏胸腺内ウイルス接種による家鶏肉腫の電子顕微鏡的観察	171
175. 新保幸太郎, 成松英明, 東 浩, 西田一巳, 室谷光三: ウイルス性腫瘍の本態に関する研究 (第3報) 家鶏肉腫各系の組織像の比較	172
176. 高橋泰常, 石沢 実, 高橋哲也: 千葉系家鶏肉腫ウイルスに関する生化学的研究	173
177. 春日 孟, 白須泰彦, 宮本博泰: Friend 病の組織学的電子顕微鏡的研究	173
178. 大橋望彦: Friend Virus Leukemia の脾から Toxohormone の分離について	175
179. 螺良義彦, 市場邦通, 木村郁夫, 松本恭一郎: Moloney 白血病ウイルスによる ddO および dd ₁ 系マウスにおける淋巴性腫瘍の発生, ウイルスの回収ならびに子孫への伝達経路	176
180. 螺良義彦, 北村 旦, 市場邦通, 木村郁夫, 丸上昌男, 松本恭一郎, 仲沢政雄: Moloney 白血病ウイルス感染マウスの形態学的研究	177
181. 松井敬介, 森脇昭介, 渡辺健太郎: C ₃ H 系, SL 系および ddN 系マウス乳癌の電子顕微鏡的研究, とくに Virus 粒子の増殖形式について	178
182. 浜崎幸雄, 村上元正: 白血病性 AKR マウスより分離した agent 接種による C ₃ H マウス乳癌誘発について	179
183. 都合により中止	
184. 宮沢政栄, 西川正夫, 牛尾暉夫, 松村茂夫, 杉山 清, 上野 陽, 山本 正, 西岡久寿弥, 織田 昭, 衣川満水: E.D. Virus による制癌の臨床的, 実験的研究 (第4報) とくに肝癌の治療に関する研究	180

VIII. 診 断

185. 神前武和, 上住南八男, 長谷川伸一, 中川信哉, 小谷宜丸, 村木敬司: Malignolipin に関する研究	182
186. 山形敏一, 中沢一郎: 血中 Malignolipin 証明法の検討	183
187. 溝口政澄, 小林春満: いわゆる Malignolipin の血中証明法の癌診断的意義	184
188. 服部正次, 松田 実, 森本健二: 癌親和性螢光色素 NTS に関する研究	184

189.	松本 哲: Davis 反応の本態について	185
190.	山口延男, 笹井外喜雄, 脇坂行一, 箕 守: 癌疾患における血清 M_1 分画 のボーラログラフ蛋白波について	186
191.	久保勝彦, 脇坂行一: 白血病患者血清の Mucoprotein (Mp) とその糖成分に ついて	187
192.	松原正香, 大下寿隆, 三井文夫, 原田充善: 癌の集団検診	188
193.	入江英雄, 村上見一, 添田博彬, 原田敬一郎: 胃のレントゲン集団検診	189
194.	有賀槐三, 高橋 淳, 田中昭平, 佐藤 博: 集検により発見した胃癌の手術症 例について	190
195.	久保勝彦, 笹井外喜雄, 脇坂行一, 高木康史: 胃液 Aceton 上清部に関する研 究(第1報)	190
196.	岩崎健資, 小林節昭, 赤木正信: 分割胃液の胃液 Ninhydrin 反応に関する研 究	191
197.	熱海 明, 水戸省吾: 胃液による癌の細胞診	192
198.	吉田良行, 井上 諒: 胃癌細胞診断適中率の向上を目的とする二, 三の試み	193
199.	三浦光恵, 小野慶一, 佐藤智信, 中山広信, 武田由紀夫, 木村正昭, 富樫 充, 太田忠造, 柴田 晋, 張間行直, 鳴海泰行: 直腸癌の集団検診について	194
200.	田崎勇三, 富永仁示: 肺癌の発育についての一考察	194
201.	田崎勇三, 古川一介, 宮島頌次, 古江 尚, 稲田 雍, 富永仁示, 佐藤 泰, 五味 誠, 田ヶ谷二三夫: 原発性肺癌の臨床的諸統計	195
202.	田淵 昭, 藤原 篤: 過去 6 年間にわたって行なった子宮癌集団検診の統計的 観察	196
203.	足高善雄, 磯島晋三, 八木 明, 尼木紹男, 長谷川てる子: 予防医学的見地よ り見た子宮癌早期診断機構の現況	197
204.	藤森速水, 木下 博, 野田 定: 余等の考案せるゴルボミクロスコープの使用 成績	198

IX. 放 射 線

205.	桑田次男, 安村美博, 蟹沢成好: 紫外線による果糖肉腫細胞の不活化に関する 研究	199
206.	土館松三, 長嶋恒義, 和賀井敏夫, 林 周一: 強力超音波による腹水腫瘍破壊	

実験	200
207. 乾 朝郎, 田端敏秀: レ線照射による MN 肉腫染色体切断に関する細胞学的研究	201
208. 前田 真, 西端一郎: MN 肉腫に関する細胞学的研究 (第2報) X 線の細胞分裂能におよぼす影響について	202
209. 大星章一, 篠崎達世: 腫瘍細胞の獲得性放射線耐性に関する実験的研究	202
210. 堀 啓二, 望月捨晴, 藤野保定: 放射線の腫瘍酵素形成機能におよぼす影響	203
211. 前田茂和: いわゆる, 放射線毒素 (C-X 物質) の生物学的効果	204
212. 陣内伝之助, 田中早苗, 小野正員, 岡島邦雄, 小林淳一, 桑原良知: 「OX 物質」の臨床 (第1報)	205
213. 原 一夫, 浜崎 靖, 酒井俊一: 上顎癌の X 線治療	206
214. 山下久雄, 網野三郎, 五味 誠: 舌癌の放射線治療成績	207
215. 山下久雄, 小林秀夫: 食道癌の放射線治療	208
216. 入江英雄, 鬼塚恵一郎, 安田輝三, 笠原 興: 進行した胃癌の放射線保存療法について	211
217. 梶林和久, 木村修治, 前田一憲, 前西 浩, 真鍋宮子, 伊藤信義, 泉 洋: 胃癌手術後の放射線治療	211
218. 小島 修, 渡辺命平: 子宮頸癌放射線療法の検討	212
219. 藤森速水, 山田文夫, 前田茂和, 木下 博, 森村正孝: 子宮頸癌症例におけるブラストバルトマクロの使用経験	213

X. 治 療

220. 中山恒明, 柳沢文憲, 本間康正, 鎌田忠夫, 鈴木恵之助, 滝沢延次郎, 笥 弘毅: 癌の再発防止に対する放射線外科療法による対策について (食道癌術前照射の理論的根拠)	214
221. 伊藤敏夫, 柳沢文憲, 本間康正, 鈴木恵之助, 岩塚迪雄, 鈴木博孝, 玉真俊一: 下部食道噴門癌の病理組織学的検討と遠隔成績	215
222. 梶谷 鏡, 星野智雄, 高木国夫, 中村 真: 胃癌 2343 例の統計的観察	217
223. 友田正信, 溝口政澄: 胃十二指腸良性疾患胃切除後の残胃断端癌について	218
224. 大岩俊夫, 戸田智博, 斎藤 拓, 原口幸昭: 胃癌外科における脾臓周囲の淋巴系の検討	220

225.	原 義雄, 勝田和夫, 高橋 実, 伊藤文弥, 星野恒夫: 最近 5 カ年間の当教室 における原発性肺癌 75 例の臨床的観察	220
226.	永井良治, 仙石光彦, 吉田 稔, 鶴飼光雄: 末期乳癌に対する副腎外科 (術後 成績の検討)	221
227.	八木日出雄: 日本子宮癌委員会第 2 回報告	222
228.	増淵一正, 久保久光, 鈴木忠雄: 子宮断端癌	224
229.	明石勝英, 橋本正淑, 小森 昭, 山田 裕: 子宮頸癌の腹膜外リンパ節廓清なら びに腔式広汎性子宮全切除に関する研究 (第 2 報)	225
230.	樋口一成, 加藤 俊, 田沢多朗, 小川重男, 小林輝夫, 蓮田 清, 小林重高, 寺島芳輝, 田中 晃, 南雲秀晃: Krukenberg 氏腫瘍の臨床と病理	226
231.	百瀬剛一, 三橋慎一: 膀胱腫瘍の遠隔成績	227
232.	楠 隆光, 園田孝夫, 大川順正, 竹内正文: 泌尿器科領域における原発性副甲 状腺機能亢進症 (副甲状腺腺腫) の経験ならびにその臨床的研究	228

XI. 増 殖

233.	嶋津 孝, 末松俊彦: Mitotic rate と平行せる二, 三の酵素系について	230
234.	高木良三郎, 杉 養吉, 高野宏一, 金子君枝, 梅田 誠, 高岡聡子, 勝田 甫, 遠藤浩良, 榎本 宏: 組織培養による膠原質形成の研究 (第 3 報) ラッテ由 来細胞株 JTC-4 および -6 の膠原質形成について	231
235.	久留 勝, 神前五郎, 松島泰次郎, 伊藤英太郎, 小川 孝, 中村俊一: 悪性腫 瘍組織中に含まれる増殖促進物質 "Oncotrephin" その後の成績について	232
236.	久留 勝, 神前五郎, 青木行俊, 森下 智, 宇都宮健生, 渡辺博茂: ラッテ再 生肝および鶏胚に含まれる細胞分裂促進物質について	233
237.	勝田 甫, 高岡聡子, 多ヶ谷勇: サル腎臓細胞の組織培養 (第 1 報) 栄養要求 について	234
238.	高岡聡子, 勝田 甫, 金子君枝: 無蛋白培地による L 株細胞 (マウス線維芽 細胞) の研究 (第 7 報) 合成培地における Amino 酸要求 (続報)	235
239.	遠藤浩良, 若林克己, 榎本 宏, 勝田 甫, 高岡聡子: 組織培養による Hor- mone 作用の研究 (第 3 報) HeLa 株細胞 (人子宮頸癌) の leucine amino- peptidase の活性と性 Hormone の影響	236
240.	小塚貞雄: 透折培養法による HeLa 細胞の増殖因子の研究	237

241.	高野宏一, 広川康子: 数種の哺乳類細胞に対する免疫血清の細胞障害作用	238
242.	小塚貞雄, 小島清秀: HeLa 細胞における血清蛋白の役割りの電顕的研究	239
243.	河合和夫, 加藤良雄: HeLa 細胞に対する可視光線の影響について	240
244.	岡野錦弥, 永井清和, 森 陽一: HeLa 株細胞におよぼす Colchicine の影響について	241

XII. 転 移

245.	藤原二郎: 胃癌細胞の運動能について (続報)	242
246.	渡辺 巖, 森川清実, 勝井富三郎, 米田光作, 長岡 豊, 高柳 裕, 山川 真: 実験的胸膜腫瘍の移植と転移に関する研究 (各種臓器への移植について)	243
247.	佐藤春郎, 引地(川島)芳子, 橋本邦久, 高橋俊雄: 腫瘍転移の研究 (第8報) リンパ腺転移に関する実験	243
248.	斉藤武郎, 佐藤春郎: 腫瘍転移の研究 (第9報) C ₃ H マウス可移植性乳癌の 転移形成能について	244
249.	山田 喬: 癌の転移と癌細胞結合性に関する研究 (第2報) ラッテ腹水肝癌細胞 の増殖に伴う島細胞間結合性および表面荷電の変動	245
250.	吉田弘一, 渡部 恒, 山口金吾, 小泉昭雄, 間島 進: 肝転移の成立機序に関 する実験的研究	246
251.	山田 喬, 足立レオナルド, 井坂英彦: 腹水肝癌の研究 (第18報) 左心室内 移植による増殖部位の系統による差	247
252.	北村四郎, 小島国次, 土屋弘吉, 渡辺昭一: 胃癌の転移の研究 (胃癌の臨床病 理学的分類について)	248
253.	田北周平, 西島早見, 岸野泰雄, 豊田滋生, 河東 極: 消化器癌の転移, とく に転移形成機序に関する研究	250
254.	林 活次, 広瀬清市, 小川 滋, 佐藤寿昌: 胃十二指腸間の壁内通路路に関す る実験的研究	251
255.	池内 彦, 小川 滋: 直腸癌の拡り方の吟味	252
256.	小林 隆, 竹内正七, 松枝和夫, 山田耕司: 子宮頸癌手術材料 163 例について の原発巣とリンパ節転移との関係についての病理組織学的研究	253
257.	藤森速水, 笠原靖史: 子宮頸癌の骨盤内リンパ節転移に関する推計学的研究	254
258.	滝沢延次郎, 中野喜久男: 癌腫の血行性転移に関する研究	255

259.	宗像秀夫, 大川知之, 大沢正司: 血中癌細胞の細胞学的検討	256
260.	間島 進, 渡部 恒, 山口金吾, 小泉昭雄, 吉田弘一: 胃癌患者の血中腫瘍細胞出現の臨床的意義	257
261.	大西盛光, 柳沢文憲, 鎌田忠夫, 松崎 功, 川名正直, 横山哲夫: 食道癌患者の末梢流血中の癌細胞について (第2報)	257
262.	若狭一夫, 近藤 敏, 橋本邦久: 肺癌患者の血中癌細胞 (第4報)	258
263.	小林 隆, 竹内正七, 笠松達弘, 小泉 博, 松枝和夫, 吉田成彦, 山田耕司, 村上 真, 杉本 毅: 婦人科悪性腫瘍患者の流血中癌細胞の検出, とくに手術時の所属静脈中出现の問題	259
264.	長谷川俊治, 和田義夫: 種々の操作後における流血中の癌細胞について	260
265.	斎藤 宏, 大森幸夫: 流血中腫瘍細胞に関する実験的研究	261
266.	佐藤 博, 溝田 成, 徳山英太郎: 血中癌細胞に対する Nitromin の影響, ラッテ腹水腫瘍による実験的研究	262

XIII. 免 疫

267.	白淵 勇: 同種腫瘍細胞間の交叉免疫に関する研究	263
268.	白淵 勇, 菅原道義, 吉田順之助: 異種腫瘍細胞間の交叉免疫に関する研究	263
269.	磯島晋三: C_3H マウス (Jax) 乳癌発生に対する能動腫瘍免疫の効果 (補遺)	264
270.	高柳尹立, 三宅康夫: 悪性腫瘍患者血清中の異常蛋白に関する免疫学的検討	265
271.	関口守正, 岡田清資, 松倉迪雄, 石橋幸雄: 担癌宿主の血清 Properdin 値に関する研究 (第2報)	266
272.	折田薫三, 三宅一忠: 血清 Properdin と癌に関する研究 (第3報) 制癌剤投与時の血清 Properdin 低下に対する諸種薬剤の効果	268
273.	相沢 幹: 条件下ラッテに異種移植して人癌の増殖におよぼす Zymosan の影響	269
274.	徳永昭夫, 岡村 純, 貴島幸彦, 神前五郎: 正常人血清による Ehrlich 腹水癌細胞溶解機構 (第7報) 易熱性因子について	270
275.	岡村 純, 東 弘, 神前五郎: 正常人血清による Ehrlich 腹水癌細胞溶解機構 (第8報) Na ion の影響について	271
276.	武田勝男, 菊地浩吉, 板倉克明, 谷垣信行: ラッテ腹水癌における特異的抗移植抗原の細胞内所在に関する研究	272

277.	武田勝男, 辻由生子, 丸山孝士, 真木常雄: 一匹のラットの重複癌間の免疫学的相互関係	273
278.	武田勝男, 福土雄幸, 谷垣信行, 板倉克明: 抗ラット癌抗体に関する研究	274
279.	武田勝男, 兼元敏隆: 武田肉腫増殖におよぼす追加移植剔出の影響	274

XIV. 症 例

280.	小林昭夫, 大橋成一, 金子 仁: 重複癌の3例	276
281.	田中 昇, 陳 維嘉, 栗林宣雄: 広汎な皮膚毛細管腫瘍栓塞症による高度の全身性浮腫を示した胃癌の1剖検例	277
282.	京極方久, 奥津 完, 青木重久: 胃原発の癌肉腫の1例	278
283.	小林久人, 加野 敏: 胃腸管系に発生した平滑筋肉腫の2剖検例	280
284.	金子 仁, 大橋成一, 福島範子: 直腸後壁に発生した骨巨細胞腫状血管腫瘍の1生検例	281
285.	松岡規男, 相良昌徳, 浜田芳郎, 浜田政義: 大網膜より発生せる悪性腫瘍 (Mesenchymoma) に Dermatomyositis を併発せる1剖検例について	282
286.	佐野量造, 花田二徳: 後腹膜に発生した Paraganglioma の1例	283
287.	田中 昇, 陳 維嘉, 栗林宣雄: 大出血によって偶然発見された巨大な腎 Angiomyolipoma: 2例報告	284
288.	近藤義雄: 膀胱骨肉腫の1例	286
289.	菅 邦彦, 浜野 信: 原発性肥胖細胞性白血病の1剖検例	286
290.	寺尾 清: 赤白血病の1剖検例	287
291.	檜沢一夫, 乾 直道: 左乳様突起部に発生した胎児性横紋筋肉腫の1剖検例	288
292.	岡本達也, 小林延年: 鰓管原性癌の1剖検例	289

特 別 講 演

I.	Sarah E. Stewart: Polyoma Virus について	291
II.	村上忠重: 胃癌の発生 —臨床的方面—	305
III.	太田邦夫: 胃癌の発生 —病理学的方面—	313

I. Biochemistry

1. COMPARATIVE STUDY OF CONSTITUTING AMINO ACIDS OF THE NUCLEAR PROTEIN IN LUNG CARCINOMA AND NORMAL LUNG TISSUE BY MEANS OF STEIN MOORE'S COLUMN CHROMATOGRAPHY

HIDEKI MIYAZAKI, KENKICHI OHO, HIROSHI TSUKASAKI,
SEIRO SASHIDA, EIZO YOKOYAMA, AKIRA SHINODA
and JIRO IKEDA (Dept. of Surgery, Tokyo Med. College)

A Nucleus was isolated from a single lung carcinoma tissue according to the Mirsky and Pollister's citric acid procedure, which after defatting, was prepared into acetone powder, 9 mg of this was hydrolysed by 6N-hydro chloric acid, which after treatment of performic acid, was subjected to Stein Moore's IR 120, 150 cm and 150 cm column chromatography and concomitant paper chromatography. Thus the comparative analysis of the constituting amino acids between normal and cancer tissue, was performed. In the case of normal tissue the presence of Pro. and the molar increase of Thr. were conspicuous, while in cancer tissue molar increase of Asp., neutral amino acid Val. basic amino acid Lys. and Arg. aromatic amino-acid Phe. were apparent.

More-over it is noteworthy the emergence of unknown peak appears between Val. and Phe. in 150 cm column, corresponding to an unknown ninhydrin positive spot at higher area of Phe. on Phenol-BuOH in two dimensional paper chromatogram, in the cancer proteins.

On this peak, or spot "x", the possibility of a peptide was ruled out because the spot "x" showed only one spot on the same area, when it was cut out, and repaper-chromatographed after 4 days hydrolysis by 6 N-HCl.

附 議

黒田吉男：私もラットの腹肝ガン細胞からヒストンを抽出して Dowex-50 カラムでアミノ酸組成をしらべました。貴方の結果と同様塩基性アミノ酸類が正常肝より多く、バリンはむしろ少く、酸性アミノ酸はやや多い結果でした。（『生化学』に発表済）

粗材料から抽出された塩基性蛋白の収量はどちらが多いでしょうか。私どもはガンの方がやや収量大でした。

宮崎：手術時摘出せる肺癌組織 50 g より核蛋白質の収量は約 10 mg である。正常肺細胞核蛋白質の収量の方が少い。

2. ETHANOLAMINE IN NORMAL LIVER, REGENERATING LIVER AND HEPATOMA OF RAT

ATSUSHI OIKAWA and TAIJIRO MATSUSHIMA

(Institute for Cancer Research, Med. School, Osaka Univ.)

Significant differences in the ethanolamine content were detected among normal liver, regenerating liver and hepatoma of rat. The fresh tissue was homogenized in 50 per cent aqueous acetone, and the homogenate was dinitrophenylated for 3 hours at room temperature in the presence of excess NaHCO_3 . The ether extractable substances of the dinitrophenylated mixture were then separated on a column of Amberlite IRC-50 (H form) using a mixture of methyl-ethyl-ketone, tetrahydrofuran and water (3:4:13) as a solvent. A dinitrophenyl derivative was thus isolated and identified with authentic sample in respects to melting point, absorption curve and paper chromatographic behavior with two different solvent systems.

The ethanolamine content of normal liver was thus estimated to be 0.22 ± 0.13 $\mu\text{mole/g}$ wet tissue in 9 determinations. Approximately 8 times higher contents were, however, found in both hepatoma and regenerating liver (48 hours after partial hepatectomy); the hepatoma being *p*-dimethylamino-azobenzene-induced hepatoma and AH 130 ascites hepatoma.

The content of ethanolamine in regenerating liver was found to increase rapidly until two days after partial hepatectomy when the mitotic activity of the tissue is maximal, and then gradually decrease to the normal level.

The biochemical significance of this phenomenon in cell division is under investigation.

3. DEOXYNUCLEOSIDIC COMPOUNDS IN TUMOR CELLS

MASAKAZU HATANAKA, JUNKO TAKEI, YUKIO SUGINO

and OSAMU HAYAISHI

(Dept. of Med. Chemistry, Faculty of Med., Kyoto Univ.)

While the physical and chemical properties as well as the biological role of DNA macromolecule have been investigated extensively, little is known about the natural occurrence of acid soluble deoxynucleosidic compounds (DC), which may play some role in the active metabolism of DNA and cell proliferation. Deoxy-CDP-choline in sea urchin eggs, deoxy-CDP-ethanolamine in calf thymus and TDP-rhamnose in

some bacteria have been found to present as new types of deoxynucleotide derivatives. Their coenzyme-like structures and distribution strongly suggest that 'masked' DC (1) must actively participate in some biosynthetic reactions of rapidly dividing cells or cell nuclei where DNA synthesis proceeds.

We investigated DC in tumor cells by using Sugino's microbioassay technique (1), by which a minute amount of DC can be specifically determined in the presence of relatively large amounts of ribonucleosidic compounds. It was found that average of 43 m μ moles thymidine equivalent of 'masked' DC and 40m μ moles of 'simple' DC per gramm wet cells were contained in ascites tumor cells of AH 130 rat, but negligible amount of 'masked' DC in ascites serum. By treating with Mitomycin C (MC), an antibiotic from *Streptomyces caespitosus*, the amount of 'masked' DC in tumor cells was increased as much as two to three fold of that in untreated cells. Tumor-bearing liver, regenerating liver and MC treated liver contained two to three times as much amount of 'masked' DC as compared with that of untreated liver.

Acid soluble fraction of MC treated tumor cells were chromatographed on a Dowex-1 formate column. The chromatographical patterns showed that the amounts of deoxy-CDP-choline and deoxy-CDP-ethanolamine were increased and the amounts of deoxynucleoside triphosphates had no significant differences as compared with MC untreated cells. The biological significance of this observation is currently under investigation.

1. Y. Sugino, N. Sugino, R. Okazaki and T. Okazaki, *Biochim. Biophys. Acts* **40**, 417 1960.

4. SOME CHEMICAL AND IMMUNOCHEMICAL PROPERTIES OF THE PROTEINS OF RAT ASCITES HEPATOMA CELLS

YUKIYA SEKINE, AKIKO IJIMA, HIDEMATSU HIRAI
and KOTARO WARABIOKA

(Dept. of Biochemistry, School of Medicine, University of Tokyo;
Cancer Institute, Japanese Foundation for Cancer Research)

This paper deals with the attempts to find out any specific protein components which exist in tumor cells.

The proteins of AH 49 rat ascites tumor cells and livers were extracted with a buffer of pH 4.8 followed by a successive extraction with a pH 7.0 buffer. The proteins were partially purified either by ammonium fractionation or isoelectric precipitation. S₂-fraction (ascites cells) or L₅-fraction (liver), extracted at pH 4.8, is composed practically with several simple proteins which contain only the trace of sugar and nucleic acids, however S₆-fraction (ascites cell) or L₆-fraction (liver),

extractable at pH 7.0, is composed with a few nucleoproteins. Some physicochemical (electrophoresis, ultra centrifuge, spectrophotometry) and chemical analysis were performed with these fractions. Some of the results are shown as below:

	Amount in the tissues	RNA	DNA	E ₂₇₈	E ₂₆₀	E ₂₆₀ /E ₂₇₈
S ₅	12.5	trace	trace	6.63	4.65	0.70
L ₅	19.4	#	#	5.55	4.0	0.72
S ₆	17.7	1.22	0.83	27.2	40.4	1.49
L ₆	13.9	1.47	1.12	37.0	59.5	1.59

Amount in the tissues: content of nitrogen of fractions per cent of total nitrogen of the tissue

RNA, DNA: mg RNA (DNA) per mg nitrogen of a fraction

E₂₇₈, E₂₆₀: optical density of a solution of 1 mgN/ml concentration.
Optical path 10.0 mm, wave length: 278 and 260.

E₂₆₀/E₂₇₈: ratio of optical density

As shown above marked differences were observed either between S and L or between S₅ (L₅) and S₆ (L₆).

After an extensive immunization of rabbits and chickens using Freund's adjuvant against the fractions, it was successful to obtain fairly reactive precipitin antisera against S₅ or S₆ fractions (antibody titer: 32-64 times). Antisera against L₅ or L₆ were not pre-eminent. Between each antiserum and antigen (each fraction and rat serum) the obvious cross reactions were demonstrated by ordinary precipitin technique, agar diffusion method and immunoelectrophoresis. However the careful absorption of the antisera against S₅-fraction with L₅ and rat serum it gave a highly specific antisera.

This specific antisera gave the precipitin reaction only with S₅ fraction, but not at all with S₆, L₅, L₆ and rat serum. The immunoelectrophoretic analysis of S₅ with the correspondent antisera revealed about fourteen precipitin lines, however with this specific antisera it gave only one but strong line.

These facts may indicate that the AH 49 tumor cells contain a highly specific antigen of protein nature. This component is precipitated with half saturated ammonium sulfate. The electrophoretic mobility of the antigen is the same as that of α -globulin proved by the starch block electrophoresis technique. The purification of the component is under progress.

5. STUDIES ON TRYPTOPHAN PYRROLASE OF TUMOR BEARING ANIMALS

TAKASHI KAWACHI, YUTAKA OKAMURA, TOMOKAZU SUZUKI,
NORIO UESAKI, SETSURO FUJII and YUICHI YAMAMURA
(Dept. of Biochemistry, School of Medicine, Kyushu Univ.)

It has been reported from our laboratory that the basic protein isolated from tumor tissues had the activities of depressing liver catalase activity and plasma iron level.

The effect of the basic protein on the activity of liver tryptophan pyrrolase (TP), and its enzyme activity in tumor bearing animals were studied.

The liver TP activity of rats transplanted with Rhodamine sarcoma showed lower level than that of normal until 10 days after inoculation and various levels at later period. However, TP activity of rats which were adrenalectomized at 3 weeks after transplantation with sarcoma showed remarkably low level without exception.

When 20 mg of the basic protein isolated from tumor tissue were injected intraperitoneally into Wister-King strain rats, the liver TP activity decreased to one-half of the normal level at 3 hours after injection. The decrement of enzyme activity continued for 24 hours and then its activity recovered to normal level gradually. The minimum effective dose of the basic protein on TP activity was 10 mg. The basic protein did not inhibit TP activity in vitro. The adaptive increment of TP activity induced by the injection of L-tryptophan was also inhibited by the administration of the basic protein.

The basic protein isolated from normal rat liver had no effect on liver TP activity of rats even when 30 mg of the basic protein were injected.

附 議

一井昭五：1) 数年前われわれは同様の実験を行い報告している。その際癌移植後期にその活性の上昇すること、および適応的に Induction させてもその適応的形成があまり押えられないことからこの酵素は Toxohormone の作用をうけないと結論した。この点に関しいかに考えられるか？

2) 演者は担癌動物の Adrenalectomy によって TP が Normal Adrenalectomized の動物ものより低下していることからこの低下を Toxohormone 作用と考えられておられるようだが、担癌動物では Corticoid の level が増加していると考えられる。TP は Induction の後は前よりも活性度がしばらくはずっと低い level にあることをわれわれは見ている。担癌動物が Adrenalectomy によって Adrenalectomized の正常動物よりも低い活性を示すのは高い Corticoid level よりの Adrenalectomy によるものとは考えられないか。

河内：1) 肝 TP 活性におよぼすトキソホルモンの影響については無効あるいは variable effect 等の報告があるが、われわれの方法によって得た腫瘍塩基性蛋白質によれば明らかな活性の低下を認めることができる。

お説のように基質による induction 後に肝 TP 活性の低下を私達も認めた。corticoid による induction 後にも同様現象があるかどうかについてはしらべていない。もし担癌動物についてそのような恐れがあるならば副腎摘出ラッテに塩基性蛋白質を注射して見ればよいわけで念のため私達は副腎摘出ラッテに

塩基性蛋白質を腹腔内注射してみたが注射後 1~2 時間以内に動物が死亡してしまうので副腎摘出ラットに対する塩基性蛋白質の影響をしらべることができなかった。その点、さらに工夫して検討を加えて見たいと思っている。

6. STUDIES ON THE ENZYME FORMATION IN TUMOR-BEARING RATS

SHIGERU SHIBA, ASAHARU TERAWAKI, JUNJI DOHI,
ICHIJI ITO and MINORU MIYATAKE

(Dept. of Surgery, Research Institute for Microbial Diseases, Osaka Univ.)

The authors have attended to study the enzyme formation, which will maintain the balance in metabolism, in tumor bearing rats, with a view to explain the cancer cachexia.

In the normal rat liver, tryptophan pyrrolase and threonine dehydrase activities increased about 10 fold 5 hours after administration of the substrate of each enzyme. In the rats bearing rhodamine sarcoma, however, the increment of these enzyme activities was much lower than those in the normal rats.

On the other hand, without administration of the substrates, there was little difference in these enzyme activities between the normal and tumor-bearing rats, though slightly decreased activities were seen in proportion to the weight of tumors.

It was observed that these adaptive formations of tryptophan pyrrolase and threonine dehydrase in the rat liver were also inhibited significantly by the intra-peritoneal injection of toxohormone (alcohol precipitate) extracted from the rhodamine sarcoma according to the modified method of Nakahara & Fukuoka's, whereas without the administration of the substrates there was no recognizable difference between the normal and toxohormone-injected rats.

These results indicate that the inhibition of adaptive formations of tryptophan pyrrolase and threonine dehydrase in the tumor-bearing rats may be caused primarily by the toxohormone rather than secondarily by the haemorrhage, infection or malnutrition, derived from the tumors. These inhibitions of the adaptive formation caused by toxohormone are considered to be the inhibition of enzyme formation itself or that of the mechanism of induction in enzyme adaptation.

附 議

藤井節郎：(1) 担癌動物では肝トリプトファンピロラーゼ活性は初期には低下し後期にはむしろ上昇するとの結果をわれわれの研究では示している。この結果は Knox などの報告と一致している。今の報告ではむしろ後期では低下するということであるが、後期に上昇したような例るみられたことはないか。

(2) トキシホルモンを正常ラットに注射したとき肝トリプトファンピロラーゼ活性は低下しなかったか。
芝：トキシホルモン投与動物の酵素活性は平均してみると変化を示したとはいえない。

7. ON SERUM PHOSPHOHEXOSE ISOMERASE AND PHOSPHO-GLUCOMUTASE IN CANCER PATIENTS

NATSUKI SAMEISHIMA, YOSHIO OHNO, NAOTAKA NAKAMURA,
SADAMICHI KOBAYASHI and TAKUJI SATO

(2nd Department of Surgery, School of Medicine, Hokkaido Univ.)

Changes in serum phosphohexose isomerase and phosphoglucomutase activities in cancer patients and cancer bearing animals (Yoshida sarcoma rats), especially those after treatment were studied.

The average value of serum phosphohexose isomerase (PHI) in normal rats is 22.3 ± 7.5 Bodansky unit and that of phosphoglucomutase (PGM) 48.5 ± 11.8 Bodansky unit. 3 days after inoculation of Yoshida sarcoma into the peritoneal cavity of the rat, both PHI and PGM activities showed increase. When inoculated subcutaneously, no increase in activities of both enzymes were noticed after 5 days, but 10 days after inoculation there is a noticeable increase in these enzyme activities. When the tumor is removed 5 days after inoculation, no increase in enzyme activity is noticed even after 10 days postoperatively. Administration of either Chromomycin 20 γ /kg/day or Mitomycin 500 γ /kg/day or Carzinophilin 500 μ /kg/day on Yoshida sarcoma rats were proved effective and in this group, both serum PHI and PGM activities were less than the control group.

The authors examined the serum PHI and PGM activities in 20 healthy human beings and obtained the values of 9~42 (average 24.3 ± 9.3) unit and of 20~81 (average 51.9 ± 17.5) unit respectively. In 69 cancer patients, the average serum PHI activity was 56.1 ± 48.7 u., PGM activity was 116.4 ± 89.5 u. and many of them showed increase. Inoperable patients showed remarkable increase in these enzyme activities than operable patients. In healthy adults PHI-PGM ratio is 0.48 ± 0.1 and there is no remarkable change in cancer patients, but patients with maxillary cancer or breast cancer with bone metastasis showed increase in PHI-PGM ratio. After radical operation of the cancer these enzyme activities showed decrease, but in cases with palliative operation enzyme activities in many cases are more increased postoperatively. Radiation therapy was done on the malignant tumor cases and 3 of them with sarcoma (Malignant mediastinal tumor, lymphosarcoma of the left neck and lymphosarcoma of the right groin) showed good results. The changes in the size of the tumors were closely related to those of enzyme activities in these cases.

In summary this study showed a correlation between change in serum PHI and PGM activities and growth of the tumors.

附 議

畑中正一：使用された dosi の抗生物質で phosphohexose isomerase, phosphoglucomutase を inhibit していないか。

鮫島：吉田内腫ラッテに対する Mitomycin, Carzinophylin, Chromomycin 使用群でとくに isomerase, mutase 活性に対する酵素活性の阻害は見られなかった。

8. GLYCOGEN PHOSPHORYLASE ACTIVITIES OF TUMORS, REGENERATING RAT LIVER AND SUCKLING RAT LIVER

MASAYURI INOUE and ATSUSHI OIKAWA
(Institute for Cancer Research, Med. School, Osaka Univ.)

1. The glycogen phosphorylase activity of various tumors, including ascites and solid tumors, was measured and the previous report by Nierenberg that this enzyme is considerably less active in tumors than in normal tissues was confirmed.
2. Tissues having a high rate of cell division, such as regenerating rat liver also have less phosphorylase activity than normal rat liver.

9. BEHAVIOR OF THE PROTEIN-BOUND DYE, N-DEMETHYLASE, CATALASE AND XANTHINE OXIDASE ACTIVITIES OF RAT LIVER DURING THE COURSE OF REPEATED ADMINISTRATIONS OF 3'-ME-DAB

HIROSHI TERAYAMA, TOSHIO YAMADA and MITSUO MATSUMOTO
(Department of Biophysics of Biochemistry, Faculty of Sci., Univ. Tokyo)

25 mg of 3'-Me-DAB dissolved in one ml of corn oil was administered intermittently (usually once a week) to rats for a long period with a help of a stomach tube. Two days after the last administration of the dye, the liver was removed and the protein bound dye, N-demethylase, catalase and xanthine oxidase activities of the liver were measured. The results are summarized as follows:

- 1) The amount of polar dye reaches a maximum after a few administrations of the dye, then gradually decreases and finally levels off.
- 2) The catalase activity decreases at first, reaching a minimum, and then gradually recovers to the normal level. The decrease of the catalase in the early period does not seem to be the result of the inhibition by the dye or its metabolite, but due to the loss of the enzyme itself.
- 3) The N-demethylase and xanthine oxidase seem to behave similarly to the

catalase during the course of the dye administration.

4) Then the interpretation for the enzymic behavior observed during the repeated dye administrations was suggested. After the acute toxicological damage period at the beginning, the hepatic cells survived may regenerate, and the regenerated young cells become more and more tolerant to the damaging effect by the carcinogen probably through the altered metabolic activities against the carcinogenic aminoazo dye. Finally the liver becomes resistant as a whole and no more hepatic damage is produced even by the repeated administrations of the dye, so far as the biochemical analysis indicates.

Under the condition described herewith, the hepatic damage does not go far enough and the change for the formation of the neoplastic cells seems to be rare.

(A part of the expenses was covered by a grant from Ministry of Education of Japan)

附 議

嶋津 孝: 1. 前報によると大量のアゾ色素を胃内に与えた場合の bound dye の量は数回投与で maximum に達し以後はその level を保つとのことであったが、今回の報告と若干の違いがあるのではないか。

2. 報告によるとアゾ色素胃内投与後の bound dye の形成量と N-demethylase 活性の変化は丁度 reciprocal な関係にあるが bound dye の形成に formaldehyde が関与するという考えと矛盾しないかどうか。

寺山: ① Protein-bound Dye の量の極大値は大体 Rat Liver について一定していて、この極大値は、投与を短時間 (例えば 2~3 日おきに) 数回連続投与しても変化しない (前報)。本報告は 1 週間ごとに 1 回 (25 mg) ずつ投与しているのであってこの場合は一定の極大値に達したあと次第に減少を示すものである。

② Protein-bound dye の消長曲線と Enzyme level の消長曲線が reciprocal な関係にあることは非常に重要な知見であって、このことは、初期の各種 Enzyme level の減少が単的に発癌性物質による細胞障害を反映しているものでありその障害メカニズムの一つに Protein-dye binding も含まれるであろう。後期において binding が減少するのは dye に対する liver の適応がましてきてこれを速かに解毒化するからであって、dye と protein の binding の chance が少くなることを意味すると考えればこの間の現象はよく理解されうる。

10. PROTEOLYTIC ENZYME IN EHRlich ASCITES CARCINOMA CELL

HEIJURO SAWADA, YUKIO SUGITA, SUSUMU TERATANI,
GENICHIRO MATSUO, MORIHIKO KATO, IKUO ISHIKAWA,
AKIRA IZAKI, KENJIN KAMINO, YOSHIHISA NAKAMURA,
TAKEHIKO INOUE, SHUJI IKEDA and HIROMU SAKAMOTO

(Osaka City University, Medical School, Department of Surgery)

The activity of proteolytic enzyme with Ehrlich ascites carcinoma is distinctly recognized at pH 4.5, 5.5, 7.0, 8.5, 9.5 and 10.0, histochemically, but at pH 4.2 and

10.5 slightly. And its activity was positive in cytoplasm but negative in nucleus.

Glycerin extract of Ehrlich ascites carcinoma cells analyzed casein and human serum albumin into amino acid, at pH 3.5-9.0 (pH of added buffer).

The salting out fraction of its glycerin extract was possessed of proteolytic activity in 0.4-0.7 saturated fraction. And activity was highest in 0.6 saturated fraction.

11. THE INFLUENCE OF γ_2 -GLOBULIN ON SUCCINIC ACID DEHYDROGENASE AND NUCLEIC ACID METABOLISM OF TUMOR BEARING ANIMAL

HACHIRO SATO, TETSURO TANEDA and HARUSHI OYAMA

(Dept. of Internal Medicine, Medical School, Kagoshima University)

We have confirmed that the γ_2 -globulin obtained from the serum of normal rabbit caused the degeneration of Ehrlich ascites cancer cell *in vitro*.

In this study, we have made an attempt to investigate whether the antigen against cancer cells has anti-cancerous action or not. γ_2 -globulin was separated by Nichol's ethanol extraction method from the serum of rabbit immunized with Ehrlich ascites cancer cells in the procedure patterned after Flax' method, and then lyophilized.

24 hours after subcutaneous injection of γ_2 -globulin into d-d mice, the influences of γ_2 -globulin on the succinic acid dehydrogenase activity and nucleic acid metabolism in liver, kidney and cancer cell were investigated *in vitro*. The test materials studied in this experiment were 10% homogenate of liver and kidney of normal and cancerous mouse and Ehrlich ascites cell. For the control, γ_2 -globulin obtained from normal rabbit was also investigated.

When the mice were injected with γ_2 -globulin or $\gamma_1+\beta$ globulin separated from serums of normal or immunized rabbit, the mortality was greater in the group treated with γ_2 -globulin than in the group treated with $\gamma_1+\beta$ globulin.

Anti-cancerous γ_2 -globulin did not show any influence on the healthy group but inhibited the decrease of enzyme activity in liver and kidney of cancer bearing animal. γ_2 -globulin obtained from non-treated rabbit showed smaller anti-cancerous effect than the former.

After injection of anti-cancerous γ_2 -globulin, DNA and RNA content of liver and kidney increased significantly in both of healthy and cancerous mouse, except the decrease of RNA in kidney of cancerous animal.

In cancer cells treated with anti-cancerous γ_2 -globulin, DNA content showed a decrease of 11% and RNA content showed an increase of 11% in average, as compared with non-treated cancer cell.

12. THE TRANSMETHYLATION REACTION IN SOME EXPERIMENTAL TUMORS OF RATS

HIROAKI NAKAZATO and KATUMI ROKUO

(2nd Department of Surgery, School of Medicine, Nagoya University)

Recently, it has been solved by important researches of R. Greenberg, M. Buchanan etc. that enzyme system using folic acid derivatives as co-enzyme has a close relationship with the biosynthesis of nucleic acid and protein components. Therefore, it is conceivable that such enzyme system has an intimate relationship with the speed of cell division or multiplication of malignant tumor.

In this view, at first, the content of methionine of Takeda sarcoma and Yoshida sarcoma were determined by using the bioassay medium. *Streptococcus faecalis* RATCC 8043 were used as the test organisms. Next, the enzymatic synthesis of methionine from serine and homocysteine by homogenate of experimental tumor of rats was observed. Then, using some anti-cancer agents, their inhibitory effect on the biosynthesis of methionine by homogenate of experimental tumor was investigated. The results are summarised as follows:

1. The content of methionine in experimental tumor of rats was more than that of normal liver tissues.
2. Synthesizing activity of methionine from serine and homocysteine in experimental tumor was almost as much as in normal liver tissue.
3. Aminopterin and Amethopterin, which are antimetabolite of folic acid, had an inhibitory effect on the biosynthesis of methionine by homogenate of experimental tumor as well as by homogenate of normal liver. But Carcinophilin, Azaserine and Mitomycin had no inhibitory effect.

13. STUDIES ON GLUCOSE METABOLISM OF EHRlich's MOUSE ASCITES CARCINOMA CELLS IN ASCITIC PLASM

HEIJURO SAWADA, YUKIO SUGITA, SUSUMU TERATANI,
SHUJI IKEDA, HIROMU SAKAMOTO, GENICHIRO MATSUO,
MORIHIKO KATO, IKUO ISHIKAWA, AKIRA IZAKI,
KENJIN KAMINO, YOSHIHISA NAKAMURA and TAKEHIKO INOUE

(Dept. of Surgery, Medical School, Osaka City Univ.)

The oxygen consumption of some ascites tumors was markedly depressed by the addition of glucose, but below the levels of glucose of 5 mg per cent there developed

a slight increase in oxygen consumption. The concentration of glucose in ascitic plasma was 20 mg per cent on an average.

When 30 mg per cent of glucose (Krebs-Ringer+P-Buffer Solution) was added to carcinoma cells, blister formation and change of mitochondria were observed.

14. ON OXYGEN CONSUMPTION OF LIVERS OF TUMOR BEARING ANIMALS

KAZUHIITO YAMAGISHI and EIICHI HIYAMA

(2nd Surgical Dept. Nihon Medical College)

It has been reported that, when a minimum of bivalent iron ion was added to liver tissue, a reaction was noticed in which O_2 consumption was markedly accentuated, and that this reaction was likely to be a reaction of high molecular organic substance caused by catalysis of bivalent iron and not likely to be caused by enzyme.

The authors, in order to study this reaction, used some tissues of malignant tumor which is characterized by lowering of respiration. It was found that, though, in DAB liver cancer tissue itself, a significant reduction in O_2 consumption was noted, in the livers of the other tumor bearing animals, more than normal amount of O_2 consumption was observed.

Further study of O_2 consumption was made through this reaction in such non-tumor tissues as toxic liver, anemic liver, starved liver, etc. It was confirmed, in phospho-lipid. Lastly a significant difference was noticed between the phospho-lipid contained in cancer tissue and that contained in non-cancer tissue.

15. THE EFFECT OF 4-NITROQUINALDINE N-OXIDE ON DPN METABOLISM IN TUMOR TISSUES

TEIZO TOMARU, MOCHIIHIKO OHASHI, and FUMIKO FUKUOKA

(Cancer Institute, Japanese Foundation for Cancer Research)

It has been reported from this laboratory that 4-nitroquinoline N-oxide derivatives inhibited anaerobic glycolysis of cancer cells, and that these compounds decreased DPN level of inhibited cells, as reported by Roitt as well as Holzer et al. in the case of ethyleneimine compounds. However, the cause of the DPN decreasing effect on cancer cells has not been cleared. The authors have pursued this cause recently. The material used in this experiment was 4-nitroquinaldine N-oxide which inhibited

the anaerobic glycolysis of cancer cells more strongly than other quinoline derivatives.

It was confirmed that the DPN levels in tumor tissue were 40 and 50% down 1 and 3 days respectively after the administration of quinaldine in tumor bearing mice. On the other hand, the DPN levels of the liver in these mice were not decreased after 1 day and only slightly reduced after 3 days.

The subcellular distribution of DPN level in rhodamine fibrosarcoma tissue was compared with that of quinaldine injected one with the following results: 1) The DPN amount of nuclear fraction was about 65% of the total in whole cancer cells. 2) The DPN level of nuclei decreased to 50% of the normal level after quinaldine injection, which seemed largely to account for the total DPN decrease in whole cancer cells.

By injecting quinaldine, the DPN synthesizing enzyme, which exists mostly in nuclei, also decreased, but DPN ase maintained the normal value.

From these experiments, it may be assumed that the decreasing effect on DPN level in tumor tissue of one of the derivatives of 4-nitroquinoline N-oxide is due to the decrease of DPN synthesizing enzyme.

16. STUDIES ON THE MECHANISM OF HAEMATOPROTEIN METABOLISM IN CANCER (I).

HACHIRO SATO, KAZUO YUNOKI, SHOGO OTSUJI, TATSURO HIGASHI,
MASATAKE MORIYAMA, TERUYA ICHIKI, HARUSHI OYAMA,
TETSUYA SAMESHIMA, SHUNJI MAEDA and SHIKAZO MAEDA

(Dept. of Internal Medicine, Medical School, Kagoshima University)

It is considered that the metabolic disturbance of haemoprotein in cancer maybe contribute to cancer cachexia. In order to investigate the mechanism of haemoprotein metabolism in cancer, therefore, we estimated protoporphyrin, catalase, δ -ALA, δ -ALA dehydrase, hemoglobin, blood corpuscle copper, serum copper, α -ketoglutaric acid and nucleic acids in liver, kidney, spleen, muscle, bone and bone marrow of Wistar strain rat bearing rhodamine sarcoma. Experiments were performed on the end of first, second, third and fourth week after implantation of sarcoma. The data obtained in cancerous animal were compared with that in non-cancerous animal.

1. When the homogenate of liver, spleen or kidney was incubated with glycine, succinic acid, $MgCl_2$, and aceto-acetic acid, the amount of protoporphyrin in each tissue did not show particular change on first week, decreased on second week and increased on third week, as compared with control tissues treated with the same

procedure.

2. The amount of δ -ALA increased in liver, kidney and spleen of animal bearing sarcoma than in non-cancerous animal. The activity of δ -ALA dehydrase was estimated after activating with the addition of cystein. The enzyme activity did not show particular change between the animal bearing sarcoma and the healthy.

3. When the homogenate of each tissue was incubated with δ -ALA, the amount of protoporphyrin increased remarkably in liver, kidney and spleen, and did not show particular change in muscle, bone and bone marrow. In the course of tumor implantation, catalase activity decreased gradually in liver and kidney, and showed no change in spleen.

4. The amount of copper increased in serum and did not show any change in blood corpuscle and liver during the observation of four weeks.

5. When the homogenate of liver, kidney and spleen was incubated with δ -ALA, the amount of α -keto-glutaric acid showed an increase but nucleic acid content did not show particular change.

17. NUCLEOTIDE IN THE LIVER OF TUMOR HOST

HEIJURO SAWADA, YUKIO SUGITA, SUSUMU TERATANI,
GENICHIRO MATUO, MORIHIKO KATO, IKUO ISHIKAWA,
AKIRA IZAKI, KENJIN KAMINO, TAKEHIKO INOUE,
YOSHIHISA NAKAMURA, SHUJI IKEDA and HIROSHI SAKAMOTO

(Dept. of Surgery, Osaka City University)

In order to clarify the alteration of nucleotide in the liver of tumor host and its energy metabolism, acid soluble nucleotide in the liver of tumor host was analyzed by column chromatography day by day after implantation of Ehrlich ascites carcinoma cell. 4 or 5 days after implantation, the phosphate levels generally began to decline and decrease in ADP and ATP was recognized.

附 議

畑中正一: 1) adenosine そのものの移植後の増減はどうでしたか。

2) uridinate との contaminate はないか。

3) 他の Base の nucleotides の消長は?

4) 補酵素型ヌクレオチドで何か知見は得られなかったか?

加藤: 1) AMP, ADP, ATP の Total は日を追って減少するが日がたつにしたがって mono 磷酸と di+tri 磷酸の比は大きくひらきます。(di+tri が小さくなる)。2) Uridinate の出現は正常時、担癌日数の浅い場合見られます。3) コハク酸の検討はしていません。

18. STUDIES ON THE CAPILLARY PERMEABILITY FACTOR OF EHRLICH CARCINOMA CELLS

HEIJURO SAWADA, YUKIO SUGITA, SUSUMU TERATANI,
GENICHIRO MATSUO, MORIHIKO KATO, IKUO ISHIKAWA,
AKIRA IZAKI, KENJIN KAMINO, YOSHIHISA NAKAMURA,
SHUJI IKEDA and HIROMU SAKAMOTO

(Dept. of Surgery, Medical School, Osaka City University)

The capillary permeability factor is distinctly recognized in the normal saline extract and homogenate of Ehrlich tumor. In the supernatant of ascites tumor, no capillary permeability factor was found.

However, if cancer cells were incubated at 37.5°C for 6 hours in normal saline or supernatant of ascites tumor, that factor obviously was recognized in the supernatant. The injected area of this factor shows histologically high graded edema without leucotoxie.

This factor is inhibited by ACTH and scarcely inhibited by cortisone.

The permeability of peritoneum becomes higher by injection of this factor intraperitoneally. Therefore the correlation of this factor and ascites production of ascites tumor can not be neglected.

19. FRACTIONATION OF LIVER CATALASE DEPRESSING FACTOR OF RAT ASCITES HEPATOMA 130.

(I.) PRELIMINARY REPORT

SHIN'ICHI TANAKA, HIDETOSHI SHIGEKANE, KAZUHIKO HATTORI,
YOSHIKI KATO and SHOJI SUGA

(1st Department of Internal Medicine, School of Medicine, Nagoya University)

There are many investigations by many workers concerning the liver catalase depressing substance of tumor tissue, and highly effective ones, such as peptides (T. Ono, et al., 1955, 1956, K. Yunoki, et al., 1960), nucleotide fraction (S. Nakagawa, et al., 1955) were obtained respectively.

On the other hand the fact that the activity of toxohormone fraction (W. Nakahara, and F. Fukuoka, 1948) was not affected by digestion with pepsin (W. Nakahara et al) or with 6N HCl suggests that the active principle is a low molecular substance.

As a first step of our investigation to identify this active principle, liver catalase

depressing activity of tumor cell hydrolysate of various degree was tested to establish a basis for the fractionation.

In this experiment we used ascites hepatoma 130 of rat as tumor material and dd strain male mice for the determination of liver catalase activity.

Liver catalase was measured by modified Euler-Josephson method 24 hours after the intraperitoneal administration of tumor hydrolysate.

RAH 130 cell was digested with pepsin for 48 hours and dialyzed against distilled water in a cellophane tube after neutralization with NaOH. Dialyzable material was condensed by evaporation and dried in a desiccator. Injection of 50 mg of this sample showed a moderate depression of liver catalase activity of mice.

Trypsin digest of this material lowered definitely at a dosis of 50 mg. but HCl hydrolysis raised this liver catalase lowering action a little more.

HCl hydrolysate of tumor cell and its filtrate were effective at a dosis of 20 mg.

These hydrolysates inhibited the catalase activity *in vitro* too.

Unexpectedly the filtrate of HCl hydrolysate of normal rat liver was proved to possess an almost equal activity against catalase both *in vivo* and *in vitro*

The attempt is now undertaken to fractionate the active principle of acid hydrolysate of tumor cells and the relation of the active component in this material to toxohormone and the nonspecificity in lowering the liver catalase activity will be clarified.

附 議

藤井節郎：塩酸加水分解では活性が上昇しているとのことですが、われわれが分離したトキソホルモン活性を有する塩基性蛋白質は加水分解で活性の上昇はみとめられなかった。むしろ少し低下するようであった。

加藤：塩基性蛋白についてはやっていないのでよくわからないが、腫瘍を単に蛋白分解酵素で水解したものは明らかにカタラーゼ活性を低下させる作用が弱い。分割を進めて活性部分の最小単位が何であるかが判明しなければこの問題の解答は与えられない。塩酸水解物で活性が失なわれないことは活性物質が非常に低分子であることを想像させる。

中原和郎：実験方法に少し難点があると思う。癌組織の中には種々なカタラーゼ阻害物質が含まれていて、それを抽出すれば *In vitro* でも *In vivo* でもカタラーゼ作用の depression が起る。これまでたびたび強調しているように toxohormene は *In vitro* での作用を示さない。

加藤：分割したものを水解すべき点については、その分割に全活性が含まれることが前提とされるが、その点については現在までには確かなデータはない。単なる塩酸水解物はいわゆるトキソホルモンやコッホザフトの混合物やその他不明のものが含まれているが、分割によりこれ等を分けることができれば、良いのではないかと考えており、そのように進めたいと思っている。

20. PURIFICATION AND PARTIAL CHARACTERIZATION OF CANCER TOXIN

HACHIRO SATO and KAZUO YUNOKI

(Dept. of Internal Medicine, Medical School, Kagoshima University)

The highly purified cancer toxin preparations, capable of reducing liver catalase in normal mice, was extracted from the crude cancer toxin fraction.

A crude cancer toxin fraction was extracted from the acetone dried powder of human malignant tissue by the extraction method of glacial acetic acid. This crude substance was effective in 10 mg. doses for a mouse. An attempt was made to separate the active principle from crude fraction by the column chromatography, Amberlite XE-64, H-form. In this procedure, three fractions were obtained from all malignant tissues studied and were designated as TH₁-, TH₂- and TH₃-fractions. The effective bioassay doses for these three fractions were 10 mg., 5 to 1 mg., 10mg, respectively. The most potent preparation was concentrated in the TH₂-fraction. TH₂-fraction was confirmed to be dialyzable. The non-dialyzable cancer toxin was also separated from the crude substance by the column chromatography, Amberlite XE-64, Na-form. This principle was effective in 0.5 mg. doses in mouse bioassay. The TH₂-fraction was also separated from the partial pepsin digests of non-dialyzable preparation. TH₂-fraction was freed from nucleic acid contamination, and composed of 80% protein and 20% lipid. The electrophoretic pattern of TH₂-fraction indicated near homogeneous, however, TH₂-fraction seemed to be made up of several active principles, all closely related in their physico-chemical behavior. TH₂-fraction was divided into three closely related subfractions, TH₂-A, -B and -C, on the Amberlite column by the application of prolonged elution technique. All of the fractions were active at the 1 mg. level and all fractions possessed almost equal activity. The N-terminal amino acid¹ of cancer toxin peptide was confirmed to be arginine in all of these subfractions. From the arginine content, the minimum molecular weights of these three subfractions were estimated to be: 5,700 for TH₂-A, 4,200 for TH₂-B and 6,400 for TH₂-C.

附 議

藤井節郎: Cancer Research に報告された論文で粗トキソホルモンをカラムクロマトグラフィーで分画されたとき、粗トキソホルモンの持つトキソホルモンの活性が非常に増加している。この原因についてどうお考えになっておられるかおうかがいします。

柚木: TH₂ 分画の活性上昇については、やや強いアルカリ処理を行っていたので変性を伴っているものかも知れない。注射量は Folin 反応から Albumine 量に換算してあるが、TH₂ の Tyrosine 含量が少いため、Folin 反応では実際の量的関係を表し得ぬものとする。

大橋望彦: TH₂-fraction は癌組織特異的に認められると解したが、column charge 量が normal の場合は 1/10 量であるので同じ量の charge において比較された場合での結果を知りたい。

N-末アルギニンを同定し、それより TH₂ の各々推定分子量を求めているが、TH₂ の3分画が column 溶出曲線からもわかるように分離不完全であるため、推定不能と考える。

柚木：TH₂ 分画の Subfraction は未だ完全に分離されるにいたっていない。したがって、かかる物質の末端アミノ酸、分子量を測定することは、不適当であるが、少くとも活性物質の N 末端アミノ酸が Arginine であることは推定できる。

Analytical scale column を用い、癌、非癌両組織の粗癌腫毒分画いずれも 50 mg を分析したが、非癌からは認める程度の TH₂ 分画は得られなかった。

柚木：肝カタラーゼ測定は白川氏の quick method で測定した。試験動物は AKR 系マウス、雄、生後 8 週間を用い、動物の飼育環境も、一定にしてあるので個体差はあまりないものと思う。しかし、活性の判定規準が確定することが望ましい。

21. ON THE SERIAL CHANGES OF ¹³¹I-LABELED SERUM PROTEIN INCORPORATED TO RATS BEARING DAB-INDUCED HEPATOMA

SHUN GOTOO and SHIGEYOSHI INUI

(1st Dept. of Internal Medicine, Gifu Prefectural Medical School)

Serial changes of radioactivity in protein fractions of the sera and the liver tissues were observed after intravenous injection of ¹³¹I-labeled homologous serum protein to normal rats and rats bearing DAB-induced hepatoma.

Three stages through these series were recognized; first stage from an hour to 24 hours after injection, second stage during several days following first one and third stage from 10th to 20th day. Radioactivities measured on serum protein of both rat group were very high an hour after injection, then markedly lowered in 24 hours, thereafter inclined to gradual decrease on several days and again considerably lowered after ten days. Radioactivities measured on soluble protein of hepatoma tissues were extremely lower an hour after injection but rather higher from 48 hours to 5th day than normal tissues, and thereafter became similarly lower. Slight radioactivities were also detected till 20th day after injection.

22. ZINC METABOLISM IN MALIGNANT TUMOR (I)

HISASHI YAMAGUCHI, SHIGEYUKI ISHIGAMI, ETSURO IKEDA,
TATSUAKI SAEGUSA, YASUMICHI KONO, TOSHIO TSUZUKI
and BUNGORO MACHII

(Dept. Int. Med., Research Inst. for Microbial Dis., Osaka Univ.)

To study the zinc metabolism in tumor bearing animals, the zinc concentration in

blood, various organs and tumor tissue were determined and the relationship between changes in zinc concentration and growth rate of tumor was observed. Male rats of Wistar strain (weighing about 150 gm.) were used. The tumor used was rhodamine sarcoma and transplanted subcutaneously. Animals were killed and viviparified at a definite interval subsequent to the transplantation (on the 1st, 3rd, 5th, 10th and 20th day). In each experiment 5 to 10 animals were used.

Samples were wet ashed and zinc was determined by dithizone method.

Results I. The zinc concentration of normal rats (10 cases) were as follows: erythrocytes, 10.6 γ /cc (18.5-7.3 γ /cc); plasma 1.1 γ /cc (1.45-0.82 γ /cc); liver, 27.4 γ /g (36.2-24.2 γ /g); spleen, 18.5 γ /g (23.6-15.6 γ /g); kidney, 17.4 γ /g (20.0-15.0 γ /g); and bone marrow 28.6 γ /g (33.0-23.0).

II. Zinc concentrations in tumor bearing animals: About 10 days after the transplantation the tumor grows at a great rate. From that time, moderate normochromic anemia occurs.

Zinc concentration of erythrocytes shows no remarkable changes for the first several days after the transplantation. From 5th day, it increased rapidly and reaches the maximum value (50% above the normal) on the 10th day, decreasing slightly thereafter. But it remains at a higher level than normal (30% above the normal on 20th day).

The zinc concentration of serum is elevated immediately after the transplantation (about 30%). It decreases, thereafter, to the minimum value of about 60% of the normal on the 20th day.

The zinc concentration of the liver is elevated transiently 30% above the normal on the 1st day. It returns to normal on the 3rd day. After which, it shows again gradual increase to the maximum value on the 20th day (about 40% above normal).

Zinc concentration of spleen is elevated about 15% about normal on the 5th day and remains at this level over the experimental period.

The Zinc concentration of kidney is lowered to the minimum value on the 3rd day (about 20% below normal), after that, it increases gradually to return to normal value on the 20th day.

The zinc concentration of bone marrow is lowered to the minimum value on the 3rd day (about 15% below normal). From 10th day, it increases rapidly and reaches the maximum values on the 20th day (about 30% above normal).

The zinc concentration of tumor tissue reveals 10 γ /gm on the 5th day and 17 γ /gm on the 20th day. Relative to the total tumor tissue, the zinc concentration increases at a great rate from 13.6 γ on the 10th day to 126 γ on the 20th day.

Thus, disturbances in zinc metabolism, such as increased zinc concentration of erythrocytes, liver, spleen, and bone marrow and decreased value of plasma, were observed in tumor bearing animals paralleling the growth of tumor.

In our laboratory further studies are now being carried out to elucidate the cause of these disturbances.

附 議

石館守三：臓器の Zn 量を測定する場合検体の採取を質的に一定とすることは困難なように思う。対照を何にとるか。N, または Ca などを対照として Zn を定量するような方法を取る必要があると思うがどうでしょうか。

演者：I 亜鉛定量については種々なる方法を慎重に検討した後に(一部放射性 ^{65}Zn も利用した) 先ほど報告した定量方法を用い, はば一定せる成績をえた。

臓器亜鉛量の定量に当っては血液中に存在する亜鉛の影響を除くために, 生体灌流後に定量する必要があると考える。

II 亜鉛は蛋白に結合しており, また, 栄養学上 Ca との関係が重視されてきているので今後 N の動きに関連して考察を加える予定である。

23. DISTRIBUTION OF ZINC IN THE VARIOUS TISSUES OF TUMOR BEARING ANIMALS (I) STUDIES WITH ^{65}Zn

SHUNJI TOKUOKA and TSUYOSHI FUJIWARA
(2nd Surgical Department, Yamaguchi Medical School)

Distribution of zinc in the blood of patients with malignant tumors has been formerly reported from our laboratory.

This time the hourly changes in the distribution of zinc in the various tissues of mice, in which M-N sarcoma was implanted or not, were investigated by the measurement of the concentration of ^{65}Zn and microautoradiograph, at intervals of from 2 hours to 64 hours after the subcutaneous injection of $^{65}\text{ZnCl}_2$.

The highest concentration of ^{65}Zn in the liver showed a rapid accumulation of the injected zinc into this organ, maximal at the end of 8 hours after the injection, which at all intervals the amounts of ^{65}Zn in the liver of tumor-bearing mice were always larger than that in the normal one.

From the microautoradiographic findings, the concentration of ^{65}Zn was found to be largest in the Glisson's capsule, next cytoplasm of liver cells, and smallest in the nuclei of liver cells and extracellular spaces, and a marked accumulation of ^{65}Zn into the Glisson's capsule was confirmed especially in case of the tumor bearing mice.

Conspicuous accumulation of ^{65}Zn into the tumor cells of ascites was accelerated in the lapse of time, while the concentration of ^{65}Zn in ascitic fluid was remained small.

The amount of ^{65}Zn in the blood cells of tumor bearing mice was always less than

that in the normal mice, but no significant difference could be observed concerning the amount of ^{65}Zn in blood plasma between the tumor bearing mice and the normal one, while the amount of ^{65}Zn in the whole blood of the tumor bearing mice was always less than that of the normal mice.

It is worthy of note that conspicuous accumulation of ^{65}Zn was observed in the liver, especially in the Glisson's capsule of the tumor bearing mice and in tumor cells, while the amount of ^{65}Zn in the circulating blood was always remained less than that in the normal mice.

24. A STUDY ON THE METABOLISM OF ZINC OF EHRLICH ASCITES AND SOLID TUMOR-BEARING MICE BY MEANS OF RADIOACTIVE ZINC

AKIRA UMAHARA, TAMITARO MIZUNOYA,
YOSHIO KURODA and YUICHI YAMAMURA

(Department of Biochemistry, Faculty of Medicine, Kyushu Univ.)

In order to investigate the distribution of zinc in tumor bearing mice, aqueous solutions of radioactive zinc-histidine, -methionine, and -tryptophan compounds were intraperitoneally injected to mice which were bearing Ehrlich ascites tumor or solid tumor and the following results were obtained.

1. Ascites tumor bearing mice

At 24 hours after injection of labelled compounds, the radioactivity of tumor cells due to the accumulated ^{65}Zn was greater in zinc-histidine injected mice compared with mice received other zinc compounds. After 24 hours radioactivities decreased gradually, and at the 5th day no differences were observed among three groups. Radioactivities of liver and ascites fluid (supernatant) were negligible compared with that of tumor cells.

2. Solid tumor bearing mice

Accumulation of ^{65}Zn was higher in liver cells than tumor cells. In liver no differences among three compounds were observed in c.p.m./100 mg of nitrogen of tissue. Their effective half lives were about 15 days.

In tumor cells, at 24 hours after injection, the accumulation of ^{65}Zn was the highest in zinc-tryptophan injected mice and its activity was 9-10 times more than that of two other compounds. At 15th day activities of tumor cells were the same in all three groups.

3. *In vitro* experiment

To suspension of Ehrlich ascites tumor cells in Krebs-Ringer solution, each of three labelled compounds were added, and activities of tumor cells were compared. Results were as follows,



4. Effects of x-ray irradiation

Effects of radiation on rate of uptake by Ehrlich ascites tumor cells were studied and no differences were observed in uptake of ^{65}Zn among tumor cells of mice received total body irradiation (100, 500, 1,000, 1,500 r).

25. STUDIES ON THE UP-TAKE OF ^{65}Zn BY THE LIVER OF TUMOR BEARING ANIMALS, AND REGENERATING LIVER

BIN HAGA, TAMITARO MIZUNOYA, AKIRA Umayahara,
TAKASHI KAWACHI, SETSURO FUJII and YUICHI YAMAMURA

(Department of Biochemistry, School of Medicine, Kyushu University)

Yamamura *et al.* previously reported that the uptake of ^{65}Zn by liver increased in the tumor bearing animals. In this report the mechanism of increasing of ^{65}Zn uptake by liver of tumor bearing animals and regenerating liver was studied. ^{65}Zn glycine complex was made by the method of Tupper *et al.* Ehrlich ascites tumor was transplanted subcutaneously into ddN strain mice weighing 20 g and ^{65}Zn glycine complex (0.4 μC) was injected subcutaneously into mice at 1 week after transplantation of tumor. The uptake of ^{65}Zn by the liver of mice was tested at 24, 48 and 72 hours after ^{65}Zn injection. The liver was digested with conc. H_2SO_4 , the radioactivity was measured with a scintillation counter and the total nitrogen was determined by the micro-Kjeldahl's method.

The uptake of ^{65}Zn (cpm/N 100 mg) by liver of the tumor bearing animals increased at 24, 48 and 72 hours after ^{65}Zn injection in comparison with that of normal mice.

One mg of the basic protein having toxohormone activity from the tumor tissue was injected intraperitoneally into ddN mice weighing 20 g and the effect of the basic protein on the uptake of ^{65}Zn by liver in various period was tested at 12 hours after subcutaneous injection of ^{65}Zn glycine complex. The uptake of ^{65}Zn by liver of mice increased to the same level as that of the tumor bearing mice at 24 hours after injection of the basic protein.

The increment of this uptake continued for 48 hours and the rate of uptake of ^{65}Zn recovered gradually to the normal level.

The minimum effective dose was 500 γ . When 10 mg of bovine serum albumin

was injected, the increment of the uptake of ^{65}Zn by liver was not observed under the conditions described above. These results indicated that the increment of the uptake of ^{65}Zn by the liver of tumor bearing animal was due to some toxic substance produced in tumor tissue. The increasing of the uptake of ^{65}Zn by the regenerating liver of ddN mice was also observed.

(文部省科学研究費による)

26. IRON METABOLISM IN MALIGNANT TUMOR (IV)

HISASHI YAMAGUCHI, SHIGEYUKI ISHIGAMI, TOMOHIRO KURAHORI,
SHUN'ICHI NAKANO, TAKESHI MORII, SEIZO YAMAGUCHI
and YOSHIMASA WATANABE

(Dept. Int. Med., Research Inst. for Microbial Diseases., Osaka University)

To study the iron metabolism in tumor-bearing animals, the blood picture and the iron concentration of plasma, various organs and tumor tissue, were determined in single tumor-bearing animals, parabiotic normal animals, and parabiotic animals in which one partner possesses a tumor and the other does not. Animals used were male Wistar rats weighing about 110 gm. Rhodamine sarcoma was transplanted subcutaneously. The animals were serially killed and viviperfused at a definite interval (on the 3rd, 7th, 14th and 21st day). The iron concentration of the plasma and tissue samples, which were wet ashed previously, were determined by the o-phenanthroline method. Parabiosis was performed by the method of Meyer and Bunster. In each experiment 3 to 5 cases were used.

The results obtained are as follows: I. Single tumor-bearing animals: Moderate normo- or hyperchromic anemia occurs from the 14th day. Plasma iron is decreased progressively from the 7th day to the minimum value on the 21st day (about 60% below the normal). The iron concentration of the liver shows a slight decrease for the 1st week after the transplantation. But there is a transient increase on the 14th day (about 40% above normal), decreasing thereafter to the level of the 7th day. The iron concentration of spleen decreases immediately after the transplantation (about 40% below normal) and remains at this level over the experimental period. The iron concentration of kidney shows no remarkable changes. The iron concentration of bone marrow decreases from the 7th day and reaches the minimum value on the 21st day (about 40% below normal). The iron concentration of tumor tissue increases at a great rate from 35.7 γ /tumor on the 7th day to 207 γ /tumor on the 21st day.

II. Parabiotic normal animals: The blood picture shows no remarkable changes. The plasma iron shows a slight transient increase on the 14th day, but it is lower

than the normal non-parabiotic rat. The iron concentration of liver increases gradually and reaches the maximum value on the 21st day (about 2 times as much as the initial value). The iron concentration of spleen increases, like liver iron, and reaches the maximum on the 21st day (3 times as much as the initial value). The iron concentration of kidney shows no remarkable changes. The iron concentration of bone marrow shows an increase and reaches the maximum value on the 21st day (about 3 times the initial value).

III. Parabiotic tumor bearing partners: On the 3rd day, RBC and Hb level iron are lower than those of parabiotic normal animals. The liver, spleen, and bone marrow contain 1.5 to 2 times as much iron as in the normal parabiotic animals. But the kidney iron concentration is almost equal to that of the normal parabiotic animals. The blood picture, plasma iron, and the iron concentration of liver, spleen and kidney remain at this level. The iron concentration of bone marrow, however, shows an increase and reaches the maximum on the 21st day (about 1.5 times the value of the 3rd day). The iron concentration of tumor tissue increases progressively to the maximum value of 234 γ /tumor on the 21st day.

IV. Parabiotic non tumor-bearing partners: No differences are observed between parabiotic non tumor-bearing partners and tumor-bearing partners. Thus, the disturbances of iron metabolism of tumor bearing parabiotic animals differ from those of non-parabiotic tumor-bearing animals. Further studies are now being carried out to elucidate these differences.

27. CLINICAL STUDIES ON THE CAUSES OF ANEMIA IN CANCEROUS DISEASES (II)

KIKU NAKAO, TADASHI MAEKAWA, MASAO HATTORI,
TAKEHISA WADA, TAKUO SHIRAKURA, SHUICHI MATSUMOTO,
NORIIHIKO TASHIRO and TERUAKI KAMIYAMA

(Department of Internal Medicine, School of Med., Gunma Univ.)

Among 65 patients with cancer of various origins admitted to our department, anemia was found in 57 patients on admission, of which severe anemia with 8 g or less Hb levels was found only in patients with significant bleeding.

To elucidate the mechanism involved in the development of anemia of cancer with gastrointestinal bleeding, investigations were carried out in these patients. The results obtained were as follows:

(1) Daily blood loss to G-I tract determined by Cr^{51} method ranged 5.1 to 125.8 ml in 10 patients with cancer of G-I tract. Proportionally to the amount of blood loss,

intravascular life span of erythrocytes was shortened and anemia was severe. It is noted that the life span was slightly shortened in cases without bleeding. The shortening was more marked in cancer patients than in patients with equal amount of blood loss due to hook worm infection or peptic ulcer.

(2) In ferrokinetics, both plasma Fe^{59} clearance and utilization of Fe^{59} for Hb synthesis were markedly accelerated in cancer patients with bleeding. Erythroblast counts in bone marrow smear were increased in these patients.

(3) No elevation of UIBC was seen in rather many cancer patients with bleeding, although serum iron was as low as in patients with iron deficiency anemia.

(4) Free protoporphyrin content of erythrocytes was increased in gastric cancer, but it was lower than in idiopathic iron deficiency anemia with equal levels of serum iron or anemia.

(5) Activity of Haem-synthesis accelerating factor in plasma was around normal in gastric cancer, whereas it was low in iron deficiency anemia.

These results indicate that the main cause of anemia developed in patients with cancer of G-I tract appears to be blood loss over the elevated erythropoietic activity, and that, however, the anemia may not attribute to iron deficiency only.

28. INHIBITORY EFFECT OF PLASMA AND ASCITIC FLUID FROM EHRlich ASCITES TUMOR MOUSE UPON CYTOTOXIC ACTIVITIES OF FATTY ACIDS

MOTOTAKA MURAKAMI, YASUO TOMARI, OSAMU OHARA,
YASUO YAGI and ICHIRO KAWAGISHI

(2nd Department of Internal Medicine, School of Medicine, Kanazawa University)

It was previously reported that the homogenate of small intestinal tissue obtained from normal mouse had the cytotoxic activities upon the Ehrlich ascites tumor cells (EATC) *in vitro*. Furthermore, it was ascertained that the cytotoxic activities of the homogenate were inhibited by simultaneous addition of the plasma or cell-free ascitic fluid from EATC-bearing mouse.

The present report deals with studies on the properties of the cytotoxic substances present in the homogenate of small intestinal tissue and on the inhibitory effect of the plasma or ascitic fluid upon the cytotoxic activities of these substances.

1) The cytotoxic substances could be extracted by ether-alcohol (2:1) and were found to be soluble in acetone. Further analytical procedures indicated that the cytotoxic activities of the acetone soluble fraction were chiefly due to the free fatty acids.

On the other hand, some fatty acids were employed in order to examine whether they have cytotoxic activities upon EATC in vitro. It was demonstrated that long chain unsaturated fatty acids (oleic acid etc.) had cytotoxic activities similar to those of the free fatty acids extracted from small intestinal tissue.

2) When an adequate amount of the plasma or ascitic fluid obtained from EATC-bearing mouse was present in the medium containing the tumor cells before addition of the fatty acids thereto, the cytotoxic activities of the fatty acids upon the tumor cells were completely inhibited. However, addition of the plasma or ascitic fluid to the medium containing the fatty acids with the tumor cells could not interrupt the cytotoxic activities of the fatty acids upon the tumor cells.

Inhibitory effect of the plasma or ascitic fluid on the cytotoxic activities of the fatty acids was partly reduced by heating for 30 minutes at 65°C, and by dialysis against 0.9% NaCl solution as well.

3) The experiments using I¹³¹-labelled oleic acid suggested that long chain unsaturated fatty acids were rapidly adsorbed to the tumor cells and that presence of the plasma or ascitic fluid in the medium partly disturbed the adsorption of fatty acids to the tumor cells.

II. Chemotherapy

29. AN ANTITUMOR COMBINATION THERAPY COMBINING SUBSTANCES INHIBITING THE SYNTHESIS OF DNA, RNA AND OF PROTEIN

MIHOKO ABE, YASUHIRO ANRAKU, YUKITO MASAMUNE
and DEN'ICHI MIZUNO

(Department of Chemistry National Institute of Health)

We have been engaged in obtaining an antitumor combination therapy, combining several substances, the mode of action of which are different from each other and aiming at a synergistic effect. The present report is a first trial to combine substances inhibiting specifically the synthesis of DNA, RNA and of protein.

The specific inhibition of DNA, RNA and protein synthesis was examined for 21 antitumor substances by using *E. coli*. (Jap. J. Med. Sc. & Biol. **12**, 453 (1959)). DNA inhibitor was mitomycin (MC) and RC-4 (p-phenylene bis(di-(1-aziridinyl)phosphinate)), RNA inhibitor was 4-nitroquinoline-N-oxide (4NQNO) and protein inhibitor was chloramphenicol (CM) and erythromycin (EM).

After many trials to combine these substances, an excellent synergistic effect was obtained in a combination of MC, 4NQNO and EM or CM. In case of solid type of tumor cells, an administration of MC (20 γ), 4NQNO (250 γ) and of EM (600 γ) per mouse per day for 7 days was employed (I). In case of ascitic type MC (0.5 γ), 4NQNO (10 γ) and EM (600 γ) per mouse per day for 7 days were administered (II). Another combination was RC-4 (800 γ), 4NQNO (250 γ) and EM (600 γ) per mouse per day (III). Each dosis as given alone exerted no effect.

The combination (I) revealed a remarkable synergistic effect in solid type of Ehrlich carcinoma exerting 78% inhibition. In its ascitic form the combination (II) exerted a prolongation of longevity more than 29 days. In case of solid type of S 180 the combination (I) (CM 1200 γ instead of EM) showed 72% inhibition, but in its ascitic form (II) exerted no effect. The combination (III) showed 69% inhibition in case of solid type of Ehrlich carcinoma.

This work was supported partly by the grant of Education Ministry.

30. INFLUENCE OF SOME CARCINOSTATICS ON THE ACTIVITIES OF LIVER CATALASE, XANTHINE OXIDASE AND URICASE IN NORMAL AND TUMOR BEARING MICE (XXVII, XXVIII)

KOTOBUKI HANO, HEITARO IWATA and AKIRA AKASHI

(Department of Pharmacology, Faculty of Pharmacy, Osaka University)

It has been established that the activity of liver catalase and uricase showed a lower value in tumor bearing animals, and liver xanthine oxidase was considered to play the part of rate limiting step in purine metabolism of cancer tissue.

In the present report, ddO strain mice were inoculated with Ehrlich ascites carcinoma, and comparative studies were performed whether the therapeutic effects of anticancer agents were paralleled with the recovery of lowered enzyme activities to their normal level.

The anticancer agents used were as follows:

1. Alkylating agents.

Triethylenethiophosphoramidate (Thio-TEPA, 5 mg/kg)

p-Phenylenediphosphoric acid tetraethyleneimide (RC-4, 60 mg/kg)

2,5-Bis-ethyleniminobenzoquinone-1,4 (D.Q., 1.6 mg/kg)

2. Purine antagonists.

8-Azaguanine (8-AG, 200 mg/kg)

6-Mercaptopurine (6-MP, 80 mg/kg)

2,6-Diaminopurine (2,6-DAP, 80 mg/kg)

3. Folic acid antagonists.

Aminopterin (A-PGA, 0.5 mg/kg)

Amethopterin (A-M-PGA, 3 mg/kg)

Each agent was administered intraperitoneally 24 hrs after inoculation of tumor cells for 5 days.

As the results, the lowered activity of catalase and uricase in tumor bearing mice was recovered to their normal range by the treatment of Thio-TEPA, RC-4, D.Q. and Sark., while, in the case of 6-MP and 8-AG, the rate of recovery delayed in spite of their prolongating effect of life span.

No considerable recovering effect was observed by A-PGA, A-M-PGA and 2,6-DAP on the lowered activity of these enzymes.

In normal mice, 6-MP, 8-AG inhibited both catalase and uricase activity, and the activity of catalase was also depressed by the administration of 2,6-DAP, A-PGA and A-M-PGA.

Xanthine oxidase activity, although no differences were found between normal and tumor bearing groups, was slightly accelerated by RC-4, A-PGA and A-M-PGA, but was inhibited by the treatment with 6-MP, 8-AG and 2,6-DAP.

In vitro experiments, Thio-TEPA, D.Q., RC-4, 8-AG and Sark. showed no influence on catalase activity, while 6-MP, 2,6-DAP, 6-CP, A-PGA and A-M-PGA inhibited.

Xanthine oxidase activity was inhibited by 8-AG, 6-MP, 2,6-DAP, A-PGA and A-M-PGA. Uricase activity was inhibited by 8-AG and 6-MP.

It may be concluded that lowered liver catalase and uricase activities in tumor bearing mice were not always recovered to the normal values in parallel with the therapeutic effects of the anticancer agents. (文部省科学研究費による)

附 議

石原 実: Alkylating Agents が正常動物肝 Catalase 活性に影響ないといわれたが、われわれの正常ラットに 1 kg/kg Nitromin 連続 7 日間注射した実験では Catalase 活性を顕著に抑制するように考える。なお制癌効果と Catalase 活性との関係はいかがであったか。

明 石: 1. われわれの実験量ではアルキル化剤が *in vitro* あるいは *in vivo* において肝 Catalase 活性に影響を与えないことを認めている。

2. 制癌効果と低下した肝 Catalase 活性の回復との関係については、薬物自体の作用を考慮するとその種類 (化学構造的に) により必ずしも効果と回復が平行するとは限らない。

31. ON THE RESPIRATION, GLYCOLYSIS, AND INCORPORATION OF ^{32}P INTO RNA AND DNA OF YOSHIDA SARCOMA CELLS CULTIVATED *IN VITRO*

TOSHIAKI EBINA, KAZUO SATO, MINRO WATANABE,
MASAHIRO SATO and NOBUKO OKAMURA

(The Research Institute for Tuberculosis and Leprosy, Tohoku University)

For the last five years, we have examined *in vitro* and *in vivo* the effects of several antitumor agents on the glycolysis, respiration, and incorporation of ^{32}P into RNA and DNA of tumor cells. However, it is not reasonable to compare the results obtained *in vitro* with those obtained *in vivo*, because cell proliferation cannot be expected under *in vitro* condition.

It was confirmed by the present experiment that Yoshida sarcoma cells could be cultivated in an equal amount mixture of bovine serum and either Earl's balanced saline or Krebs-Ringer bicarbonate solution each containing 0.4 per cent lactalbumine hydrolysate. For further examinations, Yoshida sarcoma cells were cultivated in an equal amount mixture of bovine serum and Earl's balanced saline for 24 hours and then suspended in an equal amount mixture of bovine serum and Krebs-Ringer bicarbonate or tris buffer for the measurement of glycolysis or respiration, respec-

tively. Rate of incorporation of ^{32}P into RNA and DNA of these cells during the glycolysis or respiration was also determined. It was confirmed that these cells retained their glycolytic and respiratory activities for three hours or longer in both media described above. Rate of incorporation of ^{32}P into RNA was greater than that into DNA, but there was no difference between the glycolytic and respiratory condition.

Besides, light- and electron-microscope observations revealed that Yoshida sarcoma cells preserved normal in appearance even after the measurement of metabolic activities for three hours in the above media. Moreover, viability of these cells collected after the measurement was proved by implantation into a rat.

Further, the effects of several agents on the glycolysis, respiration, and incorporation of ^{32}P into RNA and DNA were examined under the same conditions described above. Nitroimin, Carzinophilin, Mitomycin, Chromomycin, 6-mercaptapurine, and monoiodoacetic acid were examined here. In general, inhibitory effects of these agents upon the incorporation of ^{32}P were stronger under the glycolytic condition than under the respiratory condition. Monoiodoacetic acid or Carzinophilin inhibited the glycolysis very markedly and incorporation of ^{32}P was also strongly inhibited by either of them.

Yoshida sarcoma cells cultivated *in vitro* seems to be more advantageous for the study on metabolic features of malignant cell proliferation.

32. THE MEMBRANE POTENTIAL OF THE HUMAN CANCER AND RELATED CELLS (II) EFFECTS OF SEVERAL ANTITUMOR SUBSTANCES ON THE MEMBRANE POTENTIALS OF THE M-N SARCOMA CELLS

SHUNJI TOKUOKA, HISASHI MORIOKA and SEIJI TANAKA

(2nd Surgical Department, Yamaguchi Medical School)

The membrane potentials of the various tissue cells (myocardium, cerebral cortex, epithelium of the kidney, gastric mucous membrane and the liver) and tumor cells of M-N sarcoma, which was subcutaneously implanted in mice, were measured by the microelectrode technique, as same as that described in the last report.

M-N sarcoma cells averaged 63 mV negative, while the potentials of the myocardium cells were 66 mV negative on the average, the others 38-47 mV negative, measured during 10 minutes after the cessation of blood flow.

The acutely decreasing pattern of the potential curves with the lapse of time after obtaining the tissue from living bodies was also observed in the M-N sarcoma cells

and gastric mucous cells in mice, as same as that confirmed in the human gastric cancer and mucous cells, while in the potential curves of the cells of the kidney, liver and cerebral cortex the typical decreasing pattern with the lapse of time was not confirmed, but showing the gradually decreasing pattern.

48 hours after the application of antitumor substances to the tumor bearing mice, i.e. mercury-hematoporphyrin (0.125-1.0 mg/15 g body weight, into the caudal vein) or 8-azaguanine (0.116-1.16 mg/15 g body weight, into the gluteal muscles) the potential curves of the M-N sarcoma cells did not show any acutely decreasing pattern with the lapse of time (0-80 minutes) in all cases of various doses, but the gradually decreasing pattern from the beginning, which was similar to that observed in the normal cells of the kidney, liver and the cerebral cortex of normal mice, and the degree of the gradual decrease was almost in parallel with the increasing amount of dose.

When mercury-hematoporphyrin, 8-azaguanine, nitromin or mitomycin-C were added to the oxygenated mammalian Ringer's solution, in which the M-N sarcoma cells were soaked during the measurement, no significant changes were observed in the acutely decreasing pattern of the potential curves of the M-N sarcoma cells with the lapse of time (0-120 minutes).

33. THE OBSERVATION ON THE ELECTRON MICROSCOPIC PICTURES OF CANCER CELLS UNDER THE ACTION OF CHEMOTHERAPEUTIC AGENTS

SHUNGO OSATO, HAJIME MORI and MICHIO MORITA

(Central Laboratory of Fukushima Med. College)

In the previous paper we reported on the electron-microscopic pictures of cancer cells at their resting stage as well as in different phases of the mitosis. By the way, we have made a preliminary report on the pictures of cancer cells under the action of Citral, which we have been investigated during the last 15 years as cancer chemotherapeutic remedy with some success. In this paper we show the electron-microscopic aspect of the action of several anticancer remedies on cancer cells. The purpose of this work is, on the one hand, to see the changes of the fine structure of the cancer cells under such influence and, on the other, to get some idea, whether we can obtain some remedies to cooperate with each other in the chemotherapy of cancer.

The method of obtaining the electron-microscopic photos was described in the previous paper.

The following remedies have been used till today.

Aldehydes (Citral); Anticancer antibiotics: Sarcomycin, Carcinophylin, Chromomycin; Nitromin (Nitrogen mustard N-oxide); Azan (8-Aza-guanin), Colchicine and PCMB (Para-chlor-mercury-benzoic acid)

Results :

Before describing the results, we must first explain signs of degenerations produced in the electronic micrographs by remedies. In the previous paper we pointed out some important signs observed in the micrographs of tumor cells.

I. As to the nucleus :

1) Nuclear membrane gets somewhat thicker and increases in its electronic density, gives impression of stiffness and loses its double structure and fine pores under the action of anticancer agents.

2) The chromatin substance is seen normally rather diffusely in the nucleus. As the degeneration of the cell goes on, it precipitates very often in the inside of the nuclear membrane and aggregates irregularly in the nucleus showing colonies. Sometimes the chromatin substance is changed into coarse granules in irregular groupings.

3) Nucleolus. Normally shows an impression of a soft tape ball like the capillary glomerule of the kidney.

With the action of anticancer agents the nucleolus increases in electronic density as a whole, deprived of its tape glomerule structure. Sometimes it becomes weaker in density, especially fading in the peripheral part.

II. Cytoplasm :

Some bodies, as mitochondria, vesicles (ergostoplasmic reticulum) and granules in the cytoplasm, are quite sensitive to anticancer remedies.

1) The mitochondria changes often into a spherical form losing the double structure of its membrane. The cristae mitochondriales fall into disorder and then mitochondria as a whole changes into a sack or into a form looking as if it were a bubble or vacuole and then it disappears.

2) Vesicles and granules in the cytoplasm, which show ordinarily clear-cut figures, often lose the sharpness in contour and become destroyed and diminished in number.

3) The ground substance of the cytoplasm, which is ordinarily homogeneous and without structure, becomes coarsely granulous, usually scanty in the periphery of the cell.

4) The cell membrane, which has usually many processes like fringes, gets smooth or deprived of those processes.

All those cell elements suffer more or less under actions of anticancer agents. If we mark above mentioned changes with $- \pm + \ddagger \#$ according to the grades of changes, so we get a table as follows :

	Nucleus			Cytoplasm			
	Nuclear membrane	Chromatin	Nucleolus	Mitochondria	Vesicles and granules	Ground substance	Cell membrane
Nitromin	+	+	+	+++	+++	+++	+
Sarcomycin	±	+	±	+	+++	+	+
Carcinophyllin	+	+	+	+	+	+	+
Azan	+	+++	+	+	+++	+	+++
Chromomycin	±	+	±	+	+	+	+
P.C.M.B.	±	±	±	+++	+++	+ or +	+
Citral	+	+	+	+	+	+	+
Colchicine	irregular course of mitosis			Cytoplasm suffers less			

The table shows that the Nitromin is most poisonous to almost all the elements of the tumor cell, while azan injures most intensively the nuclear elements and PCMB damages selectively the cytoplasmic elements, and especially mitochondria. Combined use of anticancer remedies based on our electron microscopic investigation is now going on in animal experiment.

Nitromin resistant tumor cells;

We are interested to see in the micrograph the nitromin resistant tumor cells, which appear almost always in the course of application of this remedy to the ascites tumor rats. The experiment was made on Yoshida ascites sarcoma of rat. After repeated application of nitromin to the ascites tumor rats, the great number of cells undergo severe degeneration and destruction. But we find among them always a few tumor cells remaining less affected. The difference between the sensitive and the resistant cells is obvious. We can not find for the time being any difference between those resistant cells and untreated intact tumor cells.

34. HISTOPATHOLOGICAL STUDIES ON SUBCUTANEOUSLY TRANSPLANTED EXPERIMENTAL OSTEOGENIC SARCOMA

KENYA HORIE, MOTOO MAKITA and KAZUTO SATO

(Tachikawa Hospital)

The production of osteogenic sarcoma in tibia of albino rats at a fairly high rate by using ^{32}P combined with methylcholanthrene and benzpyrene with discussion on its histogenesis was previously reported.

Further, this sarcoma was successfully transplanted for successive generations into the backs of the same strain of albino rats.

The fact that the successive transplantation of this experimentally produced osteogenic sarcoma is rather easily done and without much change in the histological picture makes it suitable for use in therapeutic experiments. This tumor is clearly osteogenic from its histological pattern, however, the tendency of bone formation is weak. It is possible that the location of the transplant in the subcutaneous tissue of the back may interfere with bone formation. In place of ossification, fibrosis can be seen frequently. This also may be due to the subcutaneous condition of the transplant which promotes fibrosis only and interferes with the complete calcification of bone.

What are the changes in this tumor when various agents were acted upon it? The effect of X-ray is diffuse but foci of necrosis also exist. In the case of anticancerous drugs, foci of necrosis can be seen more clearly. The difference between the two cases is that with X-ray the tissue is radiated evenly throughout, where as with the drugs, which are transported by the circulation, the distribution is relatively uneven and thus the sarcomatous tissue is acted unequally.

Predominant causes fibrosis readily, which is probably due to interference with protein metabolism.

AAN effects normal bone, but does not appear to effect the immature sarcoma.

35. ELECTRONMICROSCOPIC STUDIES ON YOSHIDA SARCOMA AND ASCITES HEPATOMA (AH 130 AND 7974 STRAIN): EFFECTS OF NITROMIN (NITROGEN MUSTARD N-OXIDE)

SADAO SHINOSAWA and HIROMOTO YASUDA

(Department of Pathology, School of Med., Jikei University)

50 mg/kg Nitromin solutions were injected into the peritoneal cavities of the albino rats (Donryu strain) which had got the transplantation of the Yoshida sarcoma and two strains of Ascites Hepatomas (AH 130 and 7974), 4 to 5 days prior to the Nitromin injection. In AH 130 and Yoshida Sarcoma, the swelling of the mitochondria with decrease in electron density of ground substance was observed 1 to 6 hours after Nitromin injection followed by fragmentation and destruction of membrane system of mitochondria (12-24 hours). In the rats examined after 48 hours the tumor cells showed rather slight changes of mitochondria which could be interpreted as recovery from injury or that this might be a structure of remained intact or newly formed ascites tumor cells.

But, in AH 7974 which is rather resistant strain to Nitromin, only slight swelling of mitochondria could be found in those examined 6-12 hours after Nitromin injection.

Fragmentation and loss of normal shape of rough surfaced endoplasmic reticulum, decrease in number, and disappearance of RNA granules in cytoplasm and of rough surfaced endoplasmic reticulum, decrease in number of smooth surfaced endoplasmic reticulum (vesicular elements) etc. could be seen from 1 hour after Nitromin injection, reaching their maximum at 12 to 24 hours and after 48 hours they are minimum.

Changes of mitochondria and those of other cell components (R-Er, S-Er, RNA granules etc.) are not always parallel in their intensity. Among two tumors, namely AH 130 and Yoshida sarcoma, the changes described above are slightly more severe in the former.

36. STUDIES ON THE DRUG RESISTANCE OF TUMORS (I) DEVELOPMENT OF RESISTANCE TO ALKYLATING AGENTS IN ASCITES TUMORS

YOSHIO SAKURAI, HIROSHI IMAMURA, AYAKO MORIWAKI,
HIDEHIKO ISAKA and MOTOI ISHIDATE

(Iatrochemical Institute; Medical Institute of Sasaki Foundation)

Acquisition of resistance of Yoshida sarcoma on nitrogen mustard was experimentally proved by

- 1) *in vitro*-contact of the tumor cells with the drug solution,
- 2) storage in cool box (-80°C) for more than 3 weeks, and
- 3) mixed transplantation to rat peritoneal cavity with a naturally resistant strain of rat ascites hepatoma such as AH 7974.

The highest resistant grade of a Yoshida Sarcoma strain obtained by the repeated

Acquisition of Resistance by Contact with Nitrogen Mustard *in vitro*.
Yoshida sarcoma

Contact Concentration of Nitrogen Mustard (m.M)	RI* MEC <i>in vitro</i> (HN_2)
2.5×10^{-4}	1 (1)**
	2.5 (6)
	5 (3)
5×10^{-4}	2.5 (6)
	5 (1)
1×10^{-3}	2.5 (1)

* RI; Resistance Index (MEC on Resistant Tumor/MEC on Original Tumor)

** Figures in parentheses show number of experimental trials

contact *in vitro* with the drug attained to 2500 fold of that of the mother strain.

The untreated mother strain of Yoshida sarcoma obtained 2.5 to 5 fold resistance by a single contact with the drug as shown in the following table.

Rats inoculated with this minimum resistant strain (2.5 fold) gave an almost similar percent survivor diagram by treatment with nitromin as that of the untreated Yoshida sarcoma rats, while the same treatment gave a remarkable curative effect on rats bearing the mother strain of Yoshida sarcoma.

The resistance acquired by contact *in vitro* with nitrogen mustard exhibited cross-resistance against all alkylating agents, but resistance indices assayed by the different alkylating agents were not necessarily same. Especially there is proved a tendency that the drug used for the contact *in vitro* in order to induce resistance gives the highest resistance index among the related alkylating agents.

A strain of Yoshida sarcoma having 2500 fold resistance against nitrogen mustard has no resistance against mitomycin C or colchicine.

*) Detail of experiments concerning to resistance induction by mere contact *in vitro* with drugs and properties of the resistant tumor will be published in the issue of 1961 of Chemical and Pharmaceutical Bulletin.

(This investigation was supported by Grant CY-2799 from the National Cancer Institute, NIH, U.S. Public Health Service.)

37. STUDIES ON THE DRUG RESISTANCE OF TUMORS. (II) GRADE ESTIMATION OF RESISTANCE

YOSHIO SAKURAI, HIROSHI SATOH, HIROSHI IMAMURA,
AKIKO MORIWAKI and HIROKO IMAI (Iatrochemical Institute)

In the study of drug resistance of tumor cells, the most important problem is the establishment of technique of assaying grade of resistance quantitatively and exactly.

A method of estimation of resistance strength using cell culture of the corresponding tumor cells is presented in this proceeding, which is believed to be superior to those hitherto applied because of the following two reasons: 1) tumor resistance should be estimated in the condition excluding any host reactions and 2) a very high resistance can only be assayed *in vitro* on account of the toxic action of the drug of high concentration or dose which should drive animal to the instant death.

By tissue culture technique using Yoshida sarcoma cell, two methods are applied; 1) resistance index is defined as a ratio of minimum effective concentration (MEC)¹⁾ of the drug on *in vitro*-cultured resistant strain of tumor to MEC of the same drug on the mother strain of Yoshida sarcoma, and 2) a ratio of 50% growth inhibition

concentration (IC_{50}) of the drug *in vitro* growing cell population of a resistant strain to IC_{50} of the same drug on the mother strain is also used as resistance index.

In general the concentration for 50% growth inhibition of a drug is far lower than that for minimum effective concentration morphologically determined, but the indices obtained by both methods are approximately same. Of course the two indices might be different in case that a tangent of growth response line of the mother strain differs from that of the resistant strain. Therefore the resistance grades of two strains of tumor should be compared with the indices determined by the same one of the above describing methods.

Detail of experiment will be published in the issue of 1961 of Chemical and Pharmaceutical Bulletin.

- 1) M. Ishidate *et al.*: Chem. Pharm. Bull. 7, 873 (1959).

38. STUDIES ON THE DRUG RESISTANCE OF TUMORS. (III) POPULATION ANALYSIS OF DRUG-RESISTANT TUMORS

HIDEHIKO ISAKA, HIROSHI IMAMURA*, AYAKO MORIWAKI*,
and YOSHIO SAKURAI*

(The Medical Institute of Sasaki Foundation;

*Iatrochemical Institute of Pharmacological Research Foundation)

Employing the technique of single-cell transplantation, clonal sublines were established from the Yoshida ascites sarcoma and its HN_2 -resistant sublines. Each clone was examined for its grade of HN_2 -resistance by the *in vitro* method described by Sakurai *et al.*

1. The resistance of the whole cell population of the original Yoshida sarcoma, indicated by the minimum effective concentration of HN_2 added to the culture medium in the *in vitro* method, is 1.0×10^{-4} mM. This value is represented by the number 1.0. The grade of resistance of clones was indicated by its multiple proportion to the 1.0.

2. Seventeen clones were established from the original Yoshida sarcoma. The grade of resistance was 1.0 in 14 clones, and 2.5 in the remaining 3 clones. No homogeneity of the original cell population was demonstrated.

3. A resistant subline of the Yoshida sarcoma, resistance of which was caused by storage in "tumor bank" at -80°C ., was examined. The grade of resistance of the whole cell population of this tumor is 2.5, but 5.0 in its 11 clones and 2.5 in other 23 clones. It showed also a mosaic population of cells which were higher in resistance than those of the original strain.

4. Another resistant subline of the Yoshida sarcoma, caused by repeated application of high doses of HN_2 *in vitro*, was examined. Its resistance is 2,500 times greater than that of the original tumor. The grade of resistance in its clonal sublines was; 2,500 in 23 cases, 1,000 in 6 cases and 250 in one case. The resistance of this tumor, however, began to decrease and after 20 generations (animal-passages) it was found to be as low as 100 times.

5. All these observations seemed to provide a basis for the assumption that selection or competition among cells might play an important role in the event either of increase or decrease in the grade of resistance of a cell population. However, a marked increase such as 2,500 times in the drug resistance can not be explained only by the concept of selection, since the experience does not allow the possibility that this kind of cell may originally be present in the tumor.

This investigation was supported by the Grant CY-2799 from the National Cancer Institute, NIH, U.S. Public Health Service.

39. STUDIES ON THE CHEMOTHERAPY OF MALIGNANT TUMORS. EXPERIMENTAL STUDIES USING YOSHIDA SARCOMA CELLS

MIYOSHI URABE, TETSUJI MIZUKAMI, SHIRO TSUNAMURA,
SEIJI MIYAZAKI, KIICHI WATANABE and TEISUKE TACHIBANA
(Department of Surgery, School of Medicine, University of Kanazawa)

We have given 0.01-1 mg Nitromin daily for a week to the rats bearing subcutaneous nodule, ascitic tumors of the Nitromin sensitive as well as the Nitromin resistant Yoshida sarcoma, and observed the changes of survival time of the animals, size of tumor, numbers of tumor cells and mitotic cells. Furthermore, we have observed the inhibitory effects of the drugs upon the tumor development by histo-chemical method such as, DNA, alkaline and acid phosphatase staining.

In the Nitromin sensitive strain, administrating more than 0.05 mg of Nitromin showed the inhibitory effects upon tumor growth, while administrating 0.01 mg of Nitromin shortened contrariwise survival time of animals and brought about increase of tumor size. In the Nitromin resistant strain, administration of 1.0 mg Nitromin showed the inhibitory effects upon tumor growth, however, a tumor bearing animal died much earlier than non-treated one. DNA in the tumor cell was decreased and disappeared much more significantly in the sensitive strain than in the resistant strain. Alkaline and acid phosphatase activities were increased in the tissues as much intensively as the tumor cell was destroyed.

We have examined the changes of oxygen consumption of tumor cells and produc-

tion of lactic acid so as to observe difference of endogenous respiration and glycolysis between the sensitive and the resistant strains and further pursued the inhibitory effects of the various drugs upon the respiration and glycolysis of the both strains. Endogenous respiration and glycolysis appeared generally to be less vigorous in the resistant strain than in the sensitive strain. If the concentration of Nitrogen mustard and Nitromin was under 10^{-3} M, the drugs did not exhibit the inhibitory effect upon endogenous respiration of the resistant strain, however, they showed inhibition just when the concentration came to 2×10^{-3} M. Tespamin showed the same effect as Nitromin did. Mitomycin C in its low concentration did not inhibit endogenous respiration of the resistant strain, that means, the Nitromin resistant strain had cross resistance to Mitomycin C. Carzinophilin had no any difference of inhibiting endogenous respiration between the Nitromin sensitive and the resistant strains, therefore, the Nitromin resistant strains had no cross resistance to Carzinophilin.

Nitromin inhibited glycolysis of both the sensitive and the resistant strains much more than Nitrogen mustard. When the Nitromin or Nitrogen mustard was given in concentration 10^{-4} M, the Nitromin resistant strain produced almost the same amount of the lactic acid as no any drug was given to the tumor cell. If the drug was given in concentration over 10^{-3} M, slight inhibition of glycolysis occurred, showing less production of the lactic acid. Tespamin and Nitrogen mustard showed the same condition in glycolysis. Mitomycin C in concentration over 500 γ /ml did not bring any difference of glycolysis between the sensitive and the resistant strains. Carzinophilin did neither bring any difference.

According to findings of endogenous respiration and glycolysis *in vitro*, it was known that the Nitromin resistant Yoshida sarcoma cells showed cross resistance to Tespamin and Mitomycin C, while it did not show cross resistance to Carzinophilin.

40. ON THE MECHANISM OF RESISTANCE TO NITROGEN MUSTARD N-OXIDE IN ASCITES TUMORS

IWAO HIRONO (Department of Pathology, Gifu Medical College)

Sulfhydryl content of tumor cells in the original strain and subline of the Yoshida ascites sarcoma and ascites hepatoma, AH 13 and AH 7974, resistant to Nitrogen mustard N-oxide (MBAO), was determined according to the amperometric titration. No significant difference was observed in the total sulfhydryl content between resistant subline and original strain. Tumor cells in the resistant subline, however, contained a larger amount of non-protein sulfhydryl group than those in the corres-

ponding original strain. Since MBAO shows an affinity to sulfhydryl group, it seems to be most probable that tumor cells containing a large amount of non-protein sulfhydryl group are more endurable, owing to counteracting the action of the agents, than those containing a smaller amount. However, the susceptibility to MBAO of tumor cells in original strains was not correlative to the sulfhydryl content. Generally, susceptibility of tumor cells in original strains to a certain anti-tumor agent is thought to be determined by various mechanism, such as sulfhydryl content, permeability of cell membrane, metabolic pathway which is unaffected by the agents, or detoxication mechanism. Development of acquired-drug resistance to a certain anti-tumor agent is due to the selective survival of tumor cells which possess a specific resistant mechanism to the agent, and in case of MBAO non-protein sulfhydryl group plays an important part in such a specific resistant mechanism. Therefore, it may be inferred that the mechanism of acquired drugresistance is invariable being decided by the agent used, while the mechanism of natural resistance is much variable depending upon tumor strains, even to the same agent.

**41. FURTHER OBSERVATIONS ON THE DEVELOPMENT OF
RESISTANCE AGAINST ANTI-CANCER AGENTS :
ESTABLISHMENT OF NITROMIN RESISTANT ASCITES
HEPATOMA (AH-130 R) AND 6-MERCAPTOPURINE
RESISTANT ADENOCARCINOMA (CA 755/6 MPR)**

JUMMEI WATANABE and KYO KAJIWARA
(Research Laboratories, Takeda Pharmaceutical Ind., Ltd.)

Attentions have been paid recently to the development of drug-resistance in cancer cells clinically and experimentally and it is hoped to find effective compounds upon the resistant cells as one way to overcome the drug-resistancy. The present report deals with the establishment of Nitromin resistant Ascites Hepatoma, AH-130 R, and 6-Mercaptopurine resistant Adenocarcinoma, Ca 755/6 MPR, and some screening results obtained by using these resistant and susceptible ones simultaneously.

About seven millions of Nitromin susceptible AH-130 cells were incubated at 37°C for 30 minutes with Nitromin in various concentrations (5000, 500, 50, 5, and 0 mcg/1.5 ml), and 0.5 ml of the treated cells were inoculated into the abdominal cavity of every Donryu rat. Cytological changes caused characteristically by Nitromin and deaths of rat by tumor were checked. The tumor cells least susceptible to the treatment were aspirated from the host and the same procedure (*in vitro* incubation

and *in vivo* checking) was repeated. After five procedures it was found that the tumor cells increased from 5 mcg/1.5 ml to 3300 mcg/1.5 ml (expressed in *in vitro* concentration of Nitromin). The resistant cells were designated as "AH-130 R" and serially kept *in vivo*. The administration of a single dose of 10 mg/kg or seven doses of 2 mg/kg/day of Nitromin did not show any curative effects upon AH-130 R, whereas the original AH-130 was cured after the treatment. The resistancy hardly exhibited change for more than two years after the establishment.

After 7 injections of 10 mg/kg/day of 6 MP from the 8th day after transplantation, the subcutaneous nodule of Adencarcinoma 755 in C 57 Black mice retarded in growth, the diameter being about 25% of the control, but it grew again and the tumor was not susceptible to the second treatment. The re-grown nodule was transferred to a new host, and similar treatments were repeated. After four *in vivo* treatments, the tumor became resistant to the administration of 7 injections of 80 mg/kg/day of 6 MP, and it was serially kept after being designated as Ca 577/6 MPR.

The two resistant lines, "AH-130 R" and Ca 755/6 MPR, showed no different features from the original susceptible ones in the rate of take, survival days, growth rate and infiltrations, except the susceptibility to the respective chemotherapeutica. They are now being routinely used for the screening of chemotherapeutics and it has been found that AH-130 R is as susceptible as AH-130 to Chromomycin, and that it has a tendency to become more susceptible to the antibiotic when the *in vivo* treatment with the antibiotic repeated. It has been also found that aminocyclopentane carboxylic acid is effective to Ca 755, but ineffective to 755/6 MPR.

(文部省科学研究費による)

42. METABOLISM OF CANCER CELLS AND ITS RELATION TO ANTI-CANCER CHEMICALS (V)

REVERSIBILITY OF NITROMIN-RESISTANCY OF EHRlich ASCITES TUMOR AND CHANGES IN BIOLOGICAL AND BIOCHEMICAL PROPERTIES OF THE TUMOR CELLS

HIDEYUKI TSUKADA, AIKO INOUE, MIYO EZOE,

SEIKI FUJIWARA and TAMENORI ONOE

(Cancer Research Institute and Department of Pathology, Sapporo Medical School)

Ehrlich ascites tumor cells were obtained from (1) mice which had undergone 15 transplant generations with Nitromin treatment (R_1), (2) mice passed 30 generations with the treatment (R_2), (3) mice passed 15 generations with the treatment, then 15

generations without the treatment (R_{1a}), and (4) mice passed 30 generations without the treatment following 15 generations with the treatment (R_{1b}). The tumor cells of these lines were compared with those of original untreated tumor (S).

Resistancy of the tumor cells to Nitromin were in the increasing order of R_{1b} , R_{1a} , R_1 , and R_2 , and the difference in resistancy was slight between R_{1b} and S.

Endogenous respiration and oxidation of fatty acids were in the increasing order of S, R_{1b} , R_1 , R_{1a} , and R_2 , but the difference between R_{1a} and R_2 was slight. For oxidations of pyruvate, alpha-ketoglutarate, succinate and malate, and cytochrome oxidase activity, on the other hand, R_{1a} gave the greatest value among these tumors, although the percentage increase in these activities was much less than that in endogenous respiration. Anaerobic glycolysis showed the changes reversely proportional to those of endogenous respiration. Total nitrogen and RNA contents showed a change similar to that of endogenous respiration, differently from that of DNA.

In summary, it is suggested that Nitromin resistancy of Ehrlich ascites tumor is reversible so far. The increase in resistancy might result in the increase in oxidative respiration, RNA, and protein contents, and with the diminution of resistancy, the respiration turned again to the normal values. It is also noteworthy that in the early stages of the decrease in resistancy, the activities were found still to increase to some extent. Electron-microscopical findings characteristic to Nitromin resistancy or to the increase in respiration seem to be an increase in the number of small-sized mitochondria and an appearance of cytoplasmic vacuoles, which were noticeable in R_1 , R_2 , and R_{1a} tumor cells.

43. EXPERIMENTAL STUDIES ON THE TRANSFORMATION OF RESISTANCE TO MITOMYCIN C IN YOSHIDA SARCOMA (I)

AKIRA HOSHINO, SOJI KURITA, TAKESHI OKITA and KIYOJI KIMURA
(1st Dept. of Int. Med., School of Med. Nagoya Univ.)

We had reported on "Experimental Studies on the Transformation of Resistance to Alanine Nitrogen Mustard in Yoshida sarcoma" on this journal in 1958. Now, we report on the transformation of resistance to Mitomycin C (MC). DNA was extracted respectively from Mitomycin C (0.5 mg/kg) resistant and sensitive Yoshida sarcoma cells by Kay, Simmons, Dounce's method with Duponol. From now, each of them is called MC resistant DNA and sensitive DNA. Sensitive Yoshida sarcoma cells were incubated together with MC resistant DNA or sensitive DNA respectively at 37°C for 2 hours in a medium of ascites fluid and saline containing 0.01 M Sodium Citrate, and then transplanted intraperitoneally into rats. Three days after inocula-

tion, rats were given various doses of MC every day.

These rats were divided into four groups; 1) a group of rats inoculated the cells which were incubated with MC resistant DNA, 2) a group inoculated the cells incubated with sensitive DNA, 3) a group inoculated the cells incubated without DNA, 4) a group of rats which were not given MC.

Four groups were studied about decrease of cell counts, degenerative change of cells, prolongation of life span and inhibition of growth of intraperitoneal tumors.

Result; 1) In the 1st group inoculated the cells incubated with MC resistant DNA, Yoshida sarcoma cells showed the complete resistance to 0.1 mg/kg of MC and the incomplete resistance to 0.2 mg/kg. 2) Both the 2nd group and the 3rd group did not show any resistance to the given doses of MC.

Namely, the sensitive Yoshida sarcoma cells could be transformed to resistant by DNA extracted from MC resistant Yoshida sarcoma cells. On the contrary, the sensitive cells could not be transformed by sensitive DNA.

The grade of resistance obtained above experiment was about 1/5 to 2/5 that of the resistant subline from which the resistant DNA was extracted.

44. STUDY ON CROSS-RESISTANCE OF AMETHOPTERIN

HIDEO ENDO (Research Institute for Tuberculosis and Leprosy, Tohoku University)

Mouse leukemia L1210 was employed as tumor strain and was implanted in the peritoneal cavity of DBA mice.

It was known that L1210 6-MP 40 mg/kg resistant strain might be able to be established on the fifth generation of transfer and L1210 amethopterin 3.0 mg/kg resistant strain might be done on the sixth generation of transfer; and amethopterin showed higher inhibitory effect on L1210 6-MP 40 mg/kg resistant strain than on its sensitive strain, but 6-MP was not effective against L1210 strain resistant to amethopterin 3.0 mg/kg.

In order to block the cross-resistance of L1210 amethopterin resistant strain to 6-MP, the present work was tried.

Anti-tumor activities of 6-MP, 6-MP riboside, 8-azaguanine, 5-fluoro-uracil, 5-fluoro-2'-deoxyuridine and 6-azathymine have been tested on the strains as follows: I. Sensitive strain. II. Resistant strain to amethopterin 1.5 mg/kg. III. Resistant strain to amethopterin 1.5 mg/kg+adenine 2.0 mg/kg. IV. Resistant strain to amethopterin 1.5 mg/kg+thymine 5.0 mg/kg.

The survival time of untreated control and treated mice with 6-MP of the four

groups is shown as follows: I. 9.2-13.9; II. 11.7-16.8; III. 14.1-26.0*; IV. 14.3-20.8.

(* Three out of ten mice were still alive on the 35th day of termination of experiment.)

From the standpoint of drug-resistance it is preferable that amethopterin will be administered together with adenine.

Acknowledgment: The author wishes to express his thanks to Dr. D.J. Hutchison and Dr. C.C. Stock of Sloan-Kettering Institute for Cancer Research, New York.

45. EFFECT OF ANTI-TUMOR AND CARCINOGENIC SUBSTANCES ON DRUG RESISTANCE TRANSFORMATION IN *TRYPANOSOMA GAMBIENSE*

SHOZO INOKI and TADASUKE ONO

(Department of Parasitology, Research Institute for Microbial Diseases, Osaka University)

Since 1956, Inoki et al. have published reports about genetic problems concerning the kinetoplast- the DNA rich, self-duplicating, intracytoplasmic particle of *Trypanosoma*. The AK forms (akinetoplastic parasites) in mice increase after treatment with pararosaniline, but in mice infected with the drug-resistant strain, the AK forms do not increase. Moreover, *in vitro* the sensitive strain can be transformed into a resistant strain by DNA of the resistant strain.

The present report shows what kind of antitumor, antibiotic, carcinogenic substances and their derivatives inhibit this drug-resistance transformation.

An experimental method was carried out by mixing at least 1.0 ml of a suspension containing sensitive parasites with at least 1.0 ml of a lysate of the resistant strain plus 0.5 ml of the substances to be tested.

After standing at room temperature for 15 minutes, the mixture was inoculated into normal mice. When parasites appeared in the peripheral blood of these mice, the AK induction test was performed. This test is used to determine the grade of P-rospaniline resistance in *Trypanosoma gambiense*.

The results show that certain of the antitumor substances tested such as Mitomycin C, Actinomycin S, Sarcomycin, Sarcomycin-INH, Carzinophilin, RC4, Naramycin, Chromomycin, Nitromin, and TEM inhibited transformation in *Trypanosoma gambiense*. However azane (8-azaguanine), Colchicine, Demecolchine, had no such effect.

Also the antibiotics, Chloramphenicol, Erythromycin, Penicillin G, and Streptomycin had no such inhibition.

On the other hand, eleven carcinogenic substances, including 4-Dimethylaminoazo-

benzene, 4-Methylaminoazobenzene did inhibit the transformation.

But it was also found that 3'-Bromo-DAB, 3'-Trifluoromethyl-DAB, 4'-Nitro-DAB, 3'-Trifluoromethyl-MAB and 4'-Nitro-MAB, all derivatives of the latter two carcinogens and have no carcinogenic activity, had no inhibition. The interesting correlation between the antitumor or carcinogenic activity of these substances and their ability to inhibit this drug-resistance transformation will be studied further.

The authors wish to thank Prof. K. Fukui, Faculty of the Engineering, Kyoto University, for kindly supplying these carcinogenic substances.

46. EXPERIMENTAL STUDIES ON CHEMOTHERAPY OF MALIGNANT GROWTH EMPLOYING YOSHIDA SARCOMA ANIMALS. (XXII) ANTITUMOR ACTIVITY OF DERIVATIVES OF NITROGEN MUSTARD CONTA- INING ONLY ONE CHLOROETHYL GROUP

TOMIZO YOSHIDA, MORIZO ISHIDATE, YOSHIO SAKURAI,
KENICHI SAWATARI, HIROSHI IMAMURA,
HIROSHI SATOH and TAZUKO TASHIRO

(The Medical Institute of Sasaki Foundation; Iatrochemical Institute)

It has been well known that 3-chloropropylamino group has no biological alkylating activity contrary to the strong activity of 2-chloroethyl group. In fact, N-methyl-bis(3-chloropropyl) amine exhibits neither alkylating activity *in vitro* on thiosulfate nor antitumor effect against experimental tumors.

Recently it drew an attention of the authors that N-methyl-(2-chloroethyl)-N-(3-chloropropyl) amine showed a strong antitumor effect on Yoshida sarcoma. Nevertheless its hydrolysis and alkylation on thiosulfate in a neutral aqueous solution did not proceed over 1 molar equivalent, as anticipated from its molecular structure. In short, this compound was proved to be a monofunctional alkylating agent from the *in vitro* reaction but has the strong biological activity.

Being encouraged by this interesting observation, the following synthetic experiments were carried out.

1) Preparation of N-alkyl-N-substituted alkyl-N-2-chloroethylamines e.g. N-benzyl-N-carboxymethyl-N-2-chloroethylamine. These compounds were all inactive as anti-tumor agent, although they have a very rapid alkylating velocity.

2) Preparation of N-methyl-N-2-chloroethyl-N- ω -chloroalkylamines. These compounds are all monofunctional from the aspect of mode of reaction *in vitro* but each has a remarkable antitumor activity.

3) Preparation of N-substituted alkyl-N-2-chloroethyl-N- ω -chloro-alkylamines. In the series of these compounds, only N-substituted alkyl-N-2-chloroethyl-N-3-chloropropylamines were effective except N-benzyl-N-2-chloroethyl-6-chlorohexylamine.

Among all these compounds, N-methyl-N-2-chloroethyl-N-4-chloro-butylamine (No. 687) exhibited the greatest curative effect on Yoshida sarcoma or on rat ascites hepatomas. Its effectiveness on experimental tumors seems to be greater than that of nitromin.

The detail report will be published in the issue of 1961 of Chemical and Pharmaceutical Bulletin.

This investigation was supported by the Grant CY-2799, N.C.I., N.I.H. .

47. EXPERIMENTAL STUDIES ON CHEMOTHERAPY OF MALIGNANT GROWTH EMPLOYING YOSHIDA SARCOMA ANIMALS (XXIII). ANTITUMOR ACTION AND TOXICITY IN COMBINATION THERAPY OF NITROMIN AND N,N'-DIMETHYL-N,N'-BIS(2-CHLOROETHYL)-PIPERAZINIUM DICHLORIDE

MORIZO ISHIDATE, TOMIZO YOSHIDA, YOSHIO SAKURAI,
IWAO YAMAMOTO, MICHIKO AOSHIMA,
TAZUKO TASHIRO and HIROSHI SATOH

(Iatrochemical Institute of Pharmacological Research Foundation ;
The Medical Institute of Sasaki Foundation ; Dental College, Osaka University)

In the early stage of investigation of nitromin, an observation was noticed by S. Katsunuma that the crude product of nitromin contaminated with N,N'-dimethyl-N,N'-bis (2-chloroethyl) piperazinium dichloride (dimer) had less clinical side-effect than the pure nitromin.

From this point of view, this paper deals with the chemical, pharmacological, and pathological investigation on a composition of equimolar mixture of nitromin and dimer (nitromin-D).

Pharmacological property: Cholinergic action of nitromin on the extirpated piece of the intestine of guinea pig is almost completely suppressed by addition of 1 molar equivalent of dimer (I. Yamamoto). The action of dimer is not depending on its steric conformation (cis and trans).

Chemical properties: Velocity of hydrolysis and alkylation *in vitro* of nitromin and nitromin-D is not different, but the reduction potential of the latter is shifted about 0.15 volt to the negative by polarographic determination.

Toxicity: The toxicity on rat of nitromin-D is the same as that of nitromin (50

mg/kg, i.p.). Though maximum tolerance doses (MTD) of both two preparations on dog are not quite different (ca. 10 mg/kg. i.p.), the general physical condition of animal on administration of MTD of nitromin-D is quite better than that of nitromin.

Antitumor effect: Tumor spectrum map of nitromin-D on ascites hepatoma of rat is very resembling to that of nitromin, though there is of course a little difference in the figure of map of each tumor.

Duration of effect in the ascites *in vivo*: Nitromin-D remains effective in the ascites 120 minutes after injection (i.p.), while nitromin remains 90 minutes.

Concentration in urine and blood: Patterns of excretion in urine or progress of concentration in blood of dog after injection of nitromin-D are not different from those in case of nitromin.

Curative effect: Percent survivor diagrams of rats bearing various tumors by these two preparations were compared. As the results, there was not found much difference in curative effect of both preparations.

(文部省科学研究費, 厚生省研究費に一部よる)

48. STUDIES ON ANTITUMOR SUBSTANCES, ON O-BENZO- QUINONE ETHYLENIMINE DERIVATIVES

SEIGORO HAYASHI, HIROSHI UEKI, MORIO KAIBARA
and MASAYUKI SAKAGAWA

(Faculty of Pharmaceutical Sciences, University of Kumamoto)

1) Extracts of 110 kinds of plants and crude drugs by water, methanol, acetone and ether were examined for their anti-tumor activity by way of CAP method using Ehrlich ascites tumor cells. Several plant extracts from *Aucuba japonica*, *Trachospermum asiaticum*, *Ranunculus glaber* etc. showed marked inhibiting circle (over 40 mm.)

2) Various quinone derivatives synthesized in this laboratory were examined for their anti-tumor effect and 3,6-bisethylenimino-o-benzoquinone was most potent of the new compounds prepared. Toxicity of 3,6-bisethylenimino-o-benzoquinone is as follows:

LD_{50} = 11.6 mg/kg. (i.p.). LD_{50} (seven days) = 2.0-1.6 mg/kg/day. (i.p.). This compound also exhibits marked prolongation of survival period of mice on Ehrlich ascites tumor cells. The subcutaneous Ehrlich carcinoma was clearly retarded by peritoneal and intravenous injection of this compound.

Thus the above quinone is found to be one of the prospective anti-tumor substance with its small effective dose and relatively weak toxicity.

49. ON THE MODE OF ANTICANCER ACTION OF RC4 (IV) EFFECT OF SEVERAL ETHYLENIMINES ON OXIDATIVE PHOSPHORYLATION BY TUMOR MITOCHONDRIA

MOTOHIRO MARUYAMA (Takamine Laboratory, Sankyo Co., Ltd.)

The preceding papers of this series presented evidences that the anticancer action of RC4 is not related to its glycolysis inhibiting activity and that the same circumstances may possibly exist in the case of other ethyleneimines.

In the present report, effect of several ethyleneimines such as RC4, O-RC4, TEPA, TESPA or E 39 on oxidative phosphorylation using Ehrlich ascites tumor cell mitochondria is described.

The mitochondria was isolated by Borst's method with slight modification and suspended in 0.21 M-mannitol-0.07 M sucrose mixture. In the medium, mitochondria had maintained its phosphorylative activity fairly stable for over 12 hrs at 2° without addition of DPN.

When succinate was used as a substrate, effect of ethyleneimines was insignificant at concentration less than 1×10^{-2} M, but exceptionally O-RC4 depressed phosphorylation at concentration of 1×10^{-3} M. All of agents except RC4 preserved solution inhibited oxidation as well as phosphorylation with α -keto-glutarate as a substrate at concentration of 1×10^{-3} M or less. RC4 preserved solution showed its action only at concentration of 2×10^{-2} M or more.

In the case of pyruvate used as a substrate, similar result was obtained as in the case of α -ketoglutarate.

Oxidative phosphorylation of mouse liver mitochondria was influenced by RC4 in the same manner as that of ascites tumor mitochondria and RC4 inhibited oxidation of β -hydroxybutyrate by liver mitochondria.

From above mentioned findings, it seems that RC4 and probably other ethyleneimines inhibit pyridinenucleotide linked enzyme system rather selectively and that this inhibition may be one of causes of anticancer action of ethyleneimines.

50. STUDIES ON THE ANTITUMOR ACTIVITY OF SEVEN-MEMBERED RING STRUCTURES. (I)

MASAO ARAKAWA and YASUNOBU SATO (Takamine Laboratory)

Seven-membered ring compounds were synthesized and tested on antitumor activities in mice bearing Ehrlich ascites carcinoma. The agents were intraperi-

toneally given for five days 24 hours after transplantation of the carcinoma.

Among 14 compounds first tested, 5-nitrosotropolone (5-NT) showed antitumor activity. Derivatives of 5-NT were then synthesized and the antitumor activities were examined by determining ratio of the toxic dose to the effective dose, which varied depending upon the structure. The following compounds were found to possess antitumor activities:

Cu, Co, Zn, Mg, and Fe salts of 5-NT, 5-nitroso-3-methoxytropolone, 5-nitroso-3-formylamidotropolone, 2-isonicotinoylhydrazino-5-nitrosotropone, 2-benzeylhydrazino-5-nitrosotropone, 2-(5-nitro-2-furoyl) hydrazino-5-nitrosotropone, tropoquinone 5-ace-toxime and 5-nitrosotropolone guanidinate. Fe salt of 5-NT showed remarkable increase in activity compared with the parent compound. Studies on more derivatives of this series are in progress.

51. ANTICANCER EFFECTS OF BARBITURIC ACID, RESORCINOL, AND RHODAN DERIVATIVES ON ASCITIC AND SOLID FORMS OF EHRlich TUMOR

KOUICHI TAKANO, YASUKO HIROKAWA, YOSHIO KATO,*
DEN'ICHI MIZUNO and CHUNOSHIN UKITA*

(National Institute of Health; Faculty of Pharmaceutical Sciences,
University of Tokyo*)

Among 27 barbituric acid derivatives screened for anticancer activity using ascites as well as solid type Ehrlich carcinoma, 5-cinnamylidene (I) and 5-phenylcarbamoyl (II) barbituric acids showed a marked prolongation of longevity (more than 30 days) when administered in 1.2 mg/mouse/day for 7 days against the ascitic form, while (I) exerted no remarkable effect on the solid type. The same dosis of (II) and sodium 1-methyl-5-phenyl carbamoyl derivative (III) showed more than 75% inhibition of solid type of Ehrlich carcinoma. In the case of solid type the curative dosis was very close to LD₅₀ of the tumor bearing mice.

Thirty five resorcinol derivatives were assayed for anticancer activity through the same procedure as above. Among them octyl and decyl resorcinols showed a remarkable prolongation of longevity (more than 30 days) by administration of 1.2 mg-0.6 mg / mouse / day for 7 days. In case of solid type they did not exert good effect. On the contrary, myristyl and palmityl derivatives showed more than 50% inhibition in case of solid type, whereas they showed no effect in ascitic type. It is of interest that the optimal alkyl length is around C₈-C₁₀ for antitumor effect, while C₈ is optimal for antiascaris activity. Some additional side chains such as

nitroso at position 4 accelerated the effect against solid type, C₆, C₈ and C₁₀ derivatives exerting more than 50% inhibition.

Twenty eight aromatic rhodan derivatives, screened for anticancer activity by the same technique, were all quite toxic, but it was found that p-acylaminophenyl thiocyanate must be a possible basic structure to show antitumor activity in solid type of Ehrlich carcinoma, since the dosis of 140 γ /mouse/day for 7 days revealed more than 40% inhibition. The higher dosis could not be tested because of the scantiness of the available amount of drugs.

This work was supported partly by the grant of Education Ministry.

52. AN EXPERIMENTAL STUDY ON THE ANTINEOPLASTIC ACTION OF D-GLUCOSAMINE (I)

YOSIO WADA, NAOAKI YAMAMOTO, YUKO NAKAMURA,
SUMIO NAKANISI and MASAMICHI ONO

(The Department of Internal Medicine, Nagoya National Hospital)

In this study, the effect of d-glucosamine hydrochloride on the life span of the rats implanted with the Yoshida ascites sarcoma and on the level of agglutinin against the typhoid vaccine were observed.

1) When d-glucosamine was administered to the rats prior to the implantation of the Yoshida ascites sarcoma, the rats survived longer than those which received the agent both before and after the implantation or those which received it solely after the implantation.

2) The titers of the agglutinin against the typhoid vaccine in the rabbits which received d-glucosamine either before, after, or both before and after the injection of the typhoid vaccine were higher than control in which a saline solution was injected in place of d-glucosamine.

These results seem to indicate that d-glucosamine has an action to enhance the defense mechanism of the vital organism.

53. EFFECT OF NUCLEIC ACID PRECURSOR ON MALIGNANT NEOPLASM

UMEHARU MATSUURA (2nd. Surgical Dept., Medical School, Okayama University)

Effect of the nucleic acid precursor upon malignant neoplasm was under the experimental study of nucleic acid, enzyme and protein metabolism. Experimental mice (Strong A) were divided into two groups: one transplanted with Ehrlich ascites carcinoma intraperitoneally and the other subcutaneously.

Several drugs (Orotic acid (O-A) 4 mg/kg, Mitomycin 0.8 mg/kg, Glucuronic acid (G-A) 40 mg/kg, Thiotic acid (T-A) 4 mg/kg) were each injected into the peritoneal cavity every day after 24 hours of inoculation, and the results were obtained as follows. In O-A group the life span was prolonged about 1.4 times longer than that of the control, accumulation of ascites 1/3 to 1/4, and weight of subcutaneous tumor was 1/2.5 of the control after two weeks of inoculation.

A. Enzyme metabolism in the group inoculated intraperitoneally.

- 1) In O-A group liver catalase activity was 2.5 times higher than in the control.
- 2) In G-A and T-A groups the activities were between the two.
- 3) Kidney catalase activity showed the same results.
- 4) Liver succinic dehydrogenase activity in the control group was nearly 2 times less than that in O-A group after 10 days of inoculation, and the same in kidney succinic dehydrogenase activity.

B. Nucleic acid metabolism.

DN A-P and RN A-P were estimated by Ogur-Rosens method in 100 grams of tumors.

- 1) After two weeks of inoculation RNA showed remarkable difference, but DNA was noticeably less amount than that of the control.
- 2) Intracellular DNA, estimated microspectrophotometrically one week after intraperitoneal inoculation, was minimally depressed in the control.

C. Protein metabolism and others.

- 1) Paperelectrophoretically, total protein and albumin were improved, γ -globulin increased in O-A group.
- 2) S-GPT was slightly improved in O-A group.
- 3) In the tumor-bearing mice increase in serum calcium and decrease in sodium and potassium were more or less observed, though, no differences between O-A group and the Control were seen.
- 4) In O-A group abnormal mitoses, giant and destroyed cells were observed at an early stage.

It will be said, from the results described above, that the precursor of nucleic acid is to be used with anticancer drug, because of the suppressive effect on the malignant neoplasms.

54. STUDIES ON THE ANTI-CANCER ACTIVITIES OF AZO-COMPOUNDS

HIDEHIKO OSHIMA

(Department of Public Health, School of Medicine, Mie Prefectural University)

In the course of the studies on the ovacidal agents, it was found in our laboratory that aromatic rhodano-derivatives showed remarkable ovacidal effects against ascaris ova. Then, the screening tests for Ehrlich ascites carcinoma were carried out with a series of synthesized compounds having structural relationship. The anti-tumor effects of benzol-compounds and azo-compounds of rhodano-derivatives have been reported. About 100 azo-compounds which have no special chemically active functional group were continuously examined as to antitumor effects.

No. 68 (5-nitro, 2, 4'-dihydroxy, 2'-methylazobenzene), No. 79 (2-methyl, 4, 4'-dihydroxy, 2'-carboxyazobenzene), No. 81 (2-methyl, 5-nitro, 4'-hydroxy, 2'-carboxyazobenzene), No. 83 (2-methyl, 4, 4'-dihydroxy, 3'-carboxyazobenzene), No. 96 (1, 3-bis, (4'-hydroxy, 2'-methylphenylazo), 4-hydroxy, 6-carboxybenzen), No. 105 (1, 3-bis, (5'-nitro, 2'-methylphenylazo), 4-hydroxy, 6-carboxybenzene) and No. 220 (2, 2'-dihydroxy, 5, 5'-dinitroazobenzene) were found to be effective cytologically. No. 83 substance exhibited a life-prolongation effect.

When No. 79 substance prepared by the coupling of 5-hydroxyanthranilic acid diazonium salt with m-cresol was administered, the particularly degenerating tumor cells with pyknotic nucleus and faintly staining cytoplasm appeared on the stained preparations. However, the particularly degenerated cells could not be observed following the treatment with No. 79 substance prepared by the coupling of 4-amino-m-cresol diazonium salt with m-hydroxybenzoic acid. From these facts, it was considered that the particularly degenerating action would be due to mixed-compound in the process of synthesis. No. 79-(2) (4-methyl, 2, 4'-dihydroxy, 2'-carboxyazobenzene), one of the mixed compounds, was prepared by the coupling of 5-hydroxyanthranilic acid diazonium salt with 4-iod-m-cresol, and next by deiodonation by metallic sodium in ethylether.

When No. 79-(2) substance was treated, most of the tumor cells showed the above mentioned particular degeneration and destruction at 30 minutes after injection.

No. 79-(2) substance exhibited similarly the particular degeneration upon ascites hepatoma 7974 and Yoshida sarcoma.

55. COMBINATION CHEMOTHERAPY OF CANCER (V)

KO SATO, MIYAHIKO SATO, and TOYO NOGI

(Katsura Surgical Department, Tohoku University)

About 1,000 Osawa rats with Yoshida sarcoma were used. Spontaneous regression of tumor cells occurred in two animals, that is, 1 percent of 201 control animals. Ascitic fluid containing about 10 to 20 million tumor cells was transplanted intraperitoneally to each of a group of rats. Substances to be tested were injected subcutaneously 72 hours after tumor inoculation. We reported previously 5 of 17 animals were cured by only one injection of "crude" 2-bis(β -chloroethyl)amino-1-azaazulene. The LD_{50} and the MTD in non-tumor-bearing rats receiving subcutaneous injections of pure 2-bis(β -chloroethyl)amino-1-azaazulene were about 45 and 40 mg. per kg. of body weight, respectively. When the rats were treated with single subcutaneous injection of this compound, 20 mg., 10 mg., 5 mg., 2 mg., 1 mg., 0.5 mg., 0.2 mg., 0.1 mg., 0.05 mg., 0.02 mg. and 0.01 mg., respectively, 72 hours after tumor inoculation, none survived and they died earlier. In 1959, according to Mc Leish's vicia test method, Shimizu found radiomimetic mitoses in the root tip cell of *Vicia fava* with 0.1 percent solution of this compound, as a maximum effective dose of it. On the other hand, colchicine like pictures were found with 0.001 percent solution of the same compound, as a maximum effective dose of it. However, when the present tumor system was used, only slight changes of chromosomes in metaphase, as scattering and aggregation, appeared 3 to 24 hours after injection with the above-mentioned doses employed. Further investigation showed that this "crude" bis(β -chloroethyl)amino-1-azaazulene was a mixture of pure 2-bis(β -chloroethyl) amino-1-azaazulene and "crude" bis (β -chloroethyl) amine and 50 percent of treated animals were cured by a single subcutaneous injection of "crude" bis (β -chloroethyl) amine 40 mg. per kg. The third substance considered as N, N'-tetra (β -chloroethyl) sulfin-diamide were synthesized repeatedly in 17 times by Prof. Seto and the reproducibility of the animal data was investigated. When pure bis (β -chloroethyl) amine was combined with N, N'-tetra (β -chloroethyl) sulfindiamide, a higher ratio of cured animals was obtained in only two of 17 animal groups. Further investigation will be carried out.

(文部省科学研究費による)

56. CLINICAL STUDIES ON THE ANTICARCINOMATOUS EFFECT OF EPOXY COMPOUND UNSATURATED FATTY ACID

SATOSHI ATSUMI (Surgical Dept. of Sagamihara National Hospital)

The anticarcinomatous effect of epoxy compound unsaturated fatty acid has been studied clinically on 26 cases of the malignant tumours. The results obtained are following:

- 1) The numbers of cases have been 6 of sarcoma, 18 of earcinoma and 2 of other malignant tumours.
- 2) The dosage of this medicine was 100 mg per day, injected intramuscularly.
- 3) From my clinical observations, the apparent effect has been found in 8 cases, and a little effect in 6 cases. But no apparent effect has been found in 12 cases.
- 4) As a vital reaction of this injection, have been found fever, acceleration of the sedimentation rate, and leucocytosis. No side action was remarked.
- 5) In the cases examined pathologically and histologically, it was noticeable that this medicine had the necrotizing and softening effect to the tumours.

57. PHASE DIFFERENCE MICROSCOPIC OBSERVATION OF THE EFFECTS OF VARIOUS SURFACTANTS ON THE EHRLICH ASCITES CARCINOMA CELLS

SATOSHI OKADA, YOSHIYUKI URATSUJI, MOTOO FUKUDA,
KEN KIMURA, YOSUKE TAKAHASHI, YUKIO TAGASHIRA
and SUKEHISA HATANO

(2nd Department of Pathology, Kobe Medical College)

According to the report by Miyasaki et. al. that some kinds of organic pigment, unsoluble in water, can migrate into the Ehrlich ascites carcinoma cells with the existence of various surfactants, the effect of such surfactants themselves was studied. The surfactants used in this experiment are the products of Kaō Sekken Co. Lit. These are divided into three groups as following.

- 1) non ionic group 2) cationic group 3) anionic group

Mice of ddDK strain were prepared.

Order of the experiment are following;

- 1) Change of the Carcinoma cell count after the administration of surfactants.

- 2) Surviving days and the change of body weight.
- 3) Phase difference microscopic observation after the injection of surfactants.

Results:

- 1) Cell count decreased after the injection, and this was significant in the anionic group.
- 2) Surviving days were slightly prolonged.
- 3) By the phase difference microscope, many micro and giant cells appeared and mitochondria could be differentiated to change round form and small in size. The shape of the cells was quite variable, and some were to be degenerated. In the anionic group, cells of mitosis became fewer.

58. THE ONCOLYTIC EFFECT OF THE EXTRACT OBTAINED FROM THE STOMACH, INTESTINE, MESENTERY AND MESENTERIAL LYMPH NODES

HAYAMI KINUKAWA, KYUJI TANIKAWA,
JUNICHIRO TANAMI and HIDEO HIRANO
(Dept. Hygiene, School of Medicine, Univ. Chiba)

In our experiments of the natural resistances, it was shown that the ether-extractable substances obtained from the intestine and mesentery of mouse and guinea pig produced respiratory and multiply inhibition in Ehrlich ascites tumor and hemolysis against mammalian red cell. But, in this experiments it was employed the 40 % physical saline extract obtained from the intestine of mouse as a original material.

The hemolytic activity of the extract is not affected by heatinactivation at 56°C for 30' and at 65°C for 30'. After dialysation against 0.85% saline at 4°C for 24°, the hemolytic activity of the extract is not decreased. Also, the *in vitro* oncolytic effect of the extract is not affected by the same treatments. In the *in vitro* oncolytic experinents with tumor bearing mouse, the extract is effective.

From all parts of the intestine tract of mouse, the hemolytic substances can be obtained, but the activity titer of the extract obtained from the stomach, jejuno-ileum and mesentery is higher than from the colon. And from the intestine of mouse embryo it is not obtained but the extract of the stomach of guinea pig shows relatively high activity.

In the ether-or physical saline-extracts from the other organs of mouse, the both activity were not found.

59. CARZINOSTATIN, A NEW ANTI-TUMOR SUBSTANCE

NAKAO ISHIDA, KATSUO KUMAGAI,
KEIZO MIYAZAKI and MASASHI ITO

(Department of Bacteriology, School of Medicine, Tohoku University)

Carzinostatin was obtained from the culture filtrate of a *Streptomyces*, E-793. The filtrate, after being adjusted to pH 2.0, was treated with acid clay, and the substance was eluted with distilled water (pH 7.0). The dried substance (carzinostatin complex) was active against *S. lutea* and *B. subtilis* in the concentrations of 10-20 ug/ml, and caused degeneration of HeLa cells in the concentrations of 125-250 ug/ml. The LD₅₀ for mice was 283 mg/kg (i.v.), 137 (i.p.), and 115 (s.c.). When the complex was administered to the Ehrlich carcinoma-bearing mice (10 mg/kg/d), decrease in number of tumor cells, appearance of giant cells, destructive effect to both cellular and nuclear parts of the tumor cells were observed, and average day of survival after the successive, intraperitoneal administration for six days, started 48 hours after implantation of 3×10^6 tumor cells, was three to five times that of control. An almost complete cure was observed in 10-20% of the mice which survived over 90 days. When sarcoma 180 solid tumor was treated similarly, 70-80% inhibition by the weight of tumor was observed after two weeks, and after four weeks the tumor disappeared completely in most of the survival animals. Such an effect was particularly marked after intravenous administration; all mice survived over four weeks and 80% showed a complete cure. Similar effects were observed with Bashford carcinoma. Carzinostatin complex contains a high molecular substance (fraction A), which is positive to ninhydrin, Sakaguchi reaction, and Pauly's diazo reaction, precipitated with trichloroacetic acid, and shows a single, sharp boundary ($S=1.3$) in the ultracentrifuge. A low molecular substance (fraction B) in the complex was considered to be the main principle in explaining the antibacterial and antitumor activities of the complex. This was confirmed by observing the effect of prolonging the life of the Ehrlich carcinoma-bearing mice. However, the biological effects of the fraction B differs in various points from those of the complex, and this is now under study.

This work has been aided financially by grant from the Ministry of Education.

60. ANTI-TUMOR ACTIVITY OF THREE ANTIBIOTICS DERIVED FROM STREPTOMYCES

TADASHI ARAI and HIROYUKI HORI

(Laboratory of Antibiotics, and Lab. of Pharmacology and Toxicology, Institute of Food Microbiology, Chiba University)

Three kinds of antibiotics, designated as Nos. 120, 928 and 204 substances, were recently isolated in our laboratory and their anti-tumor activity was compared. No. 120 is orange red pigment simultaneously produced by Actinomycin source. The pigment lacks antimicrobial activity and is relatively low toxic. No. 948 was identified as Toyokamycin. No. 204 substance was obtained as a colorless crystal and proved to be chromopeptide containing quinoxaline as a chromophore. It was also revealed that the final product of No. 204 contained two kinds of crystals, melting at 140° and 210°C.

No. 120 substance strongly inhibited the growth of Rous sarcoma at a dose of 5 mg/kg/day for six days and No. 948 showed moderate inhibition at a dose of 700 mcg while No. 204 was too toxic to determine anti-tumor effect with chicken. Prolongation of survival period was also noticed with the mice bearing S180 ascitic form and treated with No. 120 substance. According to their toxicity, many mice treated with Nos. 948 and 204 substances died within 10 days. The survived, however, lived much longer than the control.

When size distribution and number of Ehrlich cancer cells treated with above three antibiotics were determined by Coulter counter, marked shift to smaller cell diameter of distribution curve was observed with No. 204 substance. The effect of No. 120 substance was not significant.

Anti-tumor cell effects were also investigated with Ehrlich cancer cells and by various staining methods. Distinct difference was not seen among these antibiotics.

61. EXPERIMENTAL STUDIES OF EFFECTS OF SYNTHETIC STEROIDS ON CANCER CELLS (III) ANTICANCER EFFECT OF STIGMASTEROL

SYUJI HOSOKAWA, KOSHI YAMASHITA, SADA O WATANABE,
KEIKO NAKAGAWA, SUGASHI NAKAYAMA, TOSHIO MISHIMA,
and NORIAKI KAMANO

(Department of Pathology, Yamaguchi Med. School)

This report deals with anticancer effects of stigmasterol, a vegetable steroid, on MN-sarcoma produced in homogeneous NA 2-strain mice. Animals were divided into the following five groups: C-control without treatment, S1-progesterone (control), S2-dehydroepiandrosterone (control), S3-stigmasteryl acetate tetrabromide, and S4-5 α -chlorostigmasteryl acetate. These steroids were given to mice subcutaneously or intramuscularly for 4 days, 250 γ once a day, either before or after transplantation of the sarcoma in the abdominal cavity. No demonstrable changes of the vaginal smears were seen with the steroid in S3 and S4. Cytomorphological studies were carried out on these animals with the following results:

1) Surviving time of the animals in groups S1 through S4 were prolonged considerably, the average being about two times of that of C, which was 7 days. When the agents were given prior to the transplantation, the surviving time was prolonged further.

2) Vigorous proliferation of the sarcoma cells was observed 4 days after the transplantation in C; the proliferation in S1-4 was approximately half as extensive. In steroid given groups, the tumor cells began to decrease in number after the 5th day of transplantation, most cells showing some degeneration.

3) In S2 and S3, the cell nucleus and cytoplasm showed a trend of enlargement when studied with supravital preparations. Cytologically, cells had degenerative changes such as swelling and vacuole formation. In contrast, a trend of decrease in size with pyknotic degeneration was seen in S1 and S4. Distribution of the nucleus cytoplasm ratios was spread on the graph in the steroid treated groups, while the distribution in C was closer and mostly near the line of ratio 1 (malignant base line).

4) Mitosis was more frequent in S1-4 than in C. However, the majority of such cells had a formation of foam-like vacuoles and the mitosis was atypical in every phase of its course. These findings suggest that stigmasterol derivatives act on tumor cells as a mitotic poison and depressant as well.

5) The degenerative changes observed after the 8th day included swelling, forma-

tion of vacuoles, many pseudopodes, swollen mitochondria, ruptured cytoplasm, nuclear edema, pyknosis, etc. The number of mitochondria varied and Janus green positive granules were enlarged and swollen. The pyknotic changes were characteristic in S1 and S4 and cell swelling was marked in S2 and S3.

6) Histologically, metastases were less extensive in S1-4 than in C. The liver cell damage was marked in S3 and S4, however.

From these cytomorphological findings, it may be concluded that stimmasterol derivatives administered to mice bearing sarcoma suppresses the development and multiplication of the tumor cells.

62. ANTI-TUMOR ACTION OF EARTHED SERA OF TUMOR-BEARING ANIMALS (XXX)

KOOZO UEDA (Dr. Saizawa's Bio-chemistry Laboratory)

この第 30 報は末梢流血を赤土で処理して得た実験結果である。これは人癌末梢血液に対して Ringer 液を添加無菌赤土の下底におき加圧 7 日間 37°C。水分は通過上昇して赤土の上部に浮上する。これを透過塩酸 Alkohol 37°C 2 時間処理し抽出液に ammonia を添加濾液に塩酸添加減圧濃縮残留物に Ringer 液を添加溶液に硫酸 ammon を添加所要の試薬を得た。これは Schlierend-band-method による電気泳動像からこの分解物を 5 mg づつ移植人癌 mouse DT-1334 の腹腔内注射を行なった場合は癌細胞の分裂停止破壊され腫瘍の発育速度が阻害縮小して 58% は効果判定上延命効果があった。末梢血液の赤土よっての分解物は抗生物質とともに Vaccine として抗癌性を有したのではないかと思われこの実験はさらに考及している。

63. STUDIES ON AN ONCOSTATIC AGENT "K.C.G." (I)

TOKIHIKO KAYAMA (Biological Institute, Wakayama Univ.)

The oncostatic agent "K.C.G." is a metabolic product of *Serratia marcescens*, containing bacterial polysaccharide. Coley, W.B. (1892) employed crude filtrates from two species of bacteria in the treatment of cancer patients. *Serratia marcescens* was one of them. Shear M.J. extracted the effective substance from the bacterial filtrate and proved that it was bacterial polysaccharide. Recently, Kayama, T. has studied

on adaptation of *Serratia marcescens* in the biological field, and succeeded to induce the polysaccharide-rich mutant strain by cadmium treatment in 1958. The metabolic product of this mutant strain of *Serratia marcescens*, the oncostatic agent "K.C.G." inhibits tumor-cell division and necrotizes tumor-tissue, and moreover, largely unaffected on normal tissue. It is an interesting problem that "K.C.G." seems to have strong effect on tumor and little effect on normal tissue. These experimental results were observed in clinical field as well as animal research.

Toxicity of "K.C.G." was extremely weak. In the case of normal mice, LD₅₀ was 4900 mg/kg by subcutaneous injection and 1175 mg/kg by intraperitoneal one. Usually, side reaction in hepatological field was not observed by "K.C.G." treatment in the case of clinical research as well as animal experiment. Therefore, it is very interesting that repeated injection of "K.C.G." may be possible for long time.

The first clinical study was made by Dr. Hishikawa, Wakayama City. He tried this agent to a severe case of metastatic bone cancer originated from breast one, in which a surprising effect was observed, both clinically and roentgenographically.

64. EXPERIMENTAL AND CLINICAL STUDIES ON THE EFFECT OF K.C.G. (I) EFFECT OF K.C.G. ON THE NORMAL RABBITS AND YOSHIDA SARCOMA

FUMITAKA SHIOMI, HIROSHI ONODA and KIYOSHI YAMAMOTO

(1st. Division of Internal Medicine, Wakayama Red-Cross Hospital)

1) Pathological findings of K.C.G. on the normal rabbits.

Various dosage 50 mg/kg, 10 mg/kg & 2 mg/kg of K.C.G. were subcutaneously injected for 2 weeks in normal rabbits. These histological findings were following:

Testis.....Atrophy of spermatic duct especially germ cells in 50 mg/kg & 10 mg/kg groups.

Thyroid gland.....Hypofunction such as diminished follicles & scanty colloid storage in 50 mg/kg group.

Lung.....Hyperemia & edema of alveolar walls in a few cases.

Another tissues.....No remarkable findings.

2) Effect of K.C.G. on Yoshida sarcoma.

a) Yoshida sarcoma cell.

50 mg/kg of K.C.G. was injected 48 hours and 72 hours after intraperitoneal inoculation. In the former, sarcoma division cells decreased about 1/3 6 hours and 1/6 24 hours after K.C.G. injection, while in the latter, the same cells decreased about

1/11 48 hours after injection and remarkable increase of neutrophilic leucocytes was seen in both cases.

b) Yoshida solid tumor.

When 20 mg/kg of K.C.G. had been injected subcutaneously for 13 days since 16 day after subcutaneous transplantation of Yoshida sarcoma cells, growth of solid tumor was arrested within few days.

c) Prolongation of life of tumor rat.

When K.C.G. was injected intraperitoneally from 12.5 mg/kg to 200 mg/kg at 48 hours after inoculation of Yoshida sarcoma cells, 1/3 rats of each groups were survived twice or more comparing with control.

67. CLINICAL EXPERIENCE IN USE OF ANTI-CANCEROUS DRUG W.T.T.C.

KINICHI NABEYA and YOSHIYUKI IJIMA

(Department of Surgery School of Medicine Chiba University)

Introduction of oriental indigent medicine "W.T.T.C." (*Wistaria chinensis*, *Terminalia chebula*, *Trapa natus*, *Coix lacryma*) and its clinical affects to the cancerous lesions were reported in 17th meeting of Japanese Cancer Society. 230 cases of cancer of the esophagus and the stomach to whom we were able to follow clinical course with use of this medication, and 342 cases of the same illness without use of this medication were comparatively studied. Cases which revealed improvement in general and physical condition as well as objective and subjective clinical symptoms were 49 cases (21%): and cases with increase in appetite 35 case, increase in weight 23 cases, improvement in bowel movements 20, diminish in abdominal discomfort 12, and improvement in spontaneous pain in 8 cases. No particular complications by intake of this medication were seen and complaints such as fatigue, pain, dysphagia or vomiting were less frequent among those who have been taking the medications. Patients who went through conservative surgical treatment such as gastrostomy, gastroenterostomy, exploratory laparotomy due to far advanced lesions, were studied with administration of this medication. 6 months survival rate following initial operation, were 32% in 23 cases with medication and 21% in 64 cases without medications in esophageal cancer.

In gastric cancer, 35% in 72 cases who were taking this medication survived and 22% in 77 cases without the medication survived. Thus the ones who where taking this medication survived longer than those without it. This medication was also given to the patients who underwent radical surgery in the esophagus and the

stomach. Rate of recurrence following surgery with the medication is 31% in 16 cases and those without them is 64% in 47 cases for the cancer of the esophagus. In gastric cancer, recurrence rate is 20% in 119 cases with medication and 37% in 154 cases without medication. Thus there is a definite sign of decrease in recurrence with those who are taking W.T.T.C.

68. SCREENING OF VARIOUS ANTI-LEUKEMIC CHEMICAL AGENTS BY OUR BONE-MARROW TISSUE CULTURE TECHNIC

KIYOSHI HIRAKI, HIROSHI SUNAMI and KOICHI KITASHIMA

(Dept. of Internal Medicine, Medical School, Okayama University)

As already reported by us at the Seventh Congress of International Hematology in Rome, in bone-marrow tissue culture of leukemia by a method devised in our laboratory leukemias demonstrate specific growth pattern, offering an important clue in the diagnosis of the disease.

Screening of various anti-leukemic chemical agents has been performed by using this technic.

First we performed bone-marrow tissue cultures of normal persons with addition of several anti-leukemic agents, such as Urethane, Nitromin, Thio-TEPA, Myleran, Demecolcin, 6-MP, 8-Azaguanine, T-431L, ACTH, Cortisone, Prednisolone, Actinomycin C, Carzinophilin, Mytomycin C and Chromomycine, in graded concentrations to the culture, and established the maximal concentration of the agents in the medium which do not impede normal bone-marrow growth.

At this maximal concentration we conducted the screening of these chemical agents for their inhibitory effects on leukemic marrow. The respective inhibitory effect of the agents on the leukemic cells is determined by evaluating the relative growth, the wandering velocity of the cells, and the cell density.

Next, in order to determine whether or not these agents selected by the screening test would inhibit platelet production by marrow megakaryocytes, we performed culture of normal bone-marrow with drugs in the medium at the same concentration.

We evaluated platelet production of megakaryocytes by the number and behavior of megakaryocytes in the growth area. As reported at the Sixth Congress of International Society of Hematology in Boston, the megakaryocyte with tentacle formation is the representative one actively producing platelets. The appearance of many such megakaryocytes in the growth area indicates active platelet formation in the bone-marrow.

In order to determine the effects of these drugs on erythropoiesis we performed

tissue culture of bone-marrow suspension. We found out that Nitromin and Thio-TEPA markedly impede both thrombopoiesis and erythropoiesis, 6-MP mildly and Myleran only slightly, while Prednisolone has no inhibitory effect.

Conclusion :

In acute myelogenous and lymphocytic leukemias Prednisolone is most effective, followed by the combination of Prednisolone and 6-MP. In monocytic leukemia the combination of Prednisolone and 6-MP is most effective. Myleran seems best in chronic myelogenous leukemia and Prednisolone in chronic lymphocytic leukemia.

69. EXPERIMENTAL AND CLINICAL STUDIES ON NITROMIN D

JUNICHI TOKUOKA, SHIGERU MIZOTA, HIROSHI SATO,
TAKUMI TSUNEMATSU, HIDETORO TOKUYAMA,
HIROSHI SATO* and TAZUKO TASHIRO*

(Kyoundo Hospital, The Medical Institute of Sasaki Foundation ;

*The Iatrochemical Institute of Pharmacological Research Foundation)

Nitromin D is composed of HN_2N -oxide and dimer. The ratio of HN_2N -oxide to dimer in the drug is 2 : 3. This new drug suppresses almost completely the cholinergic action of Nitromin on the extirpated intestines so far examined by Magnus' method. Maximal tolerated dosis of the new drug in the rat is 125 mg/kg, while 12.5 mg/kg in the dog. The dosis in each animal is just the same as that of Nitromin. However, toxic symptoms of the animal which was given the maximal tolerated dosis of Nitromin D were quite less.

Nitromin D remains effective upon the Yoshida sarcoma cells in the abdominal cavity of rats for about 2 hours after the intraperitoneal injection, while Nitromin remains effective for 90 minutes. The duration of effective dose of Nitromin D on the tumor cells, detected in the blood and the urine after the intraperitoneal application in dogs, was almost the same as that of Nitromin. Comparative examinations on the effects of these two drugs upon the Yoshida sarcoma as well as 5 different strains of the ascites hepatoma in rats revealed no marked differences in their curative effects.

In the clinical trials, 49 cases of cancer treated with Nitromin D were examined. In a case of 64-year-old man who has been suffering from cancer of the right lung and metastatic growths at the right supraclavicular region, the tumors disappeared after repeated injections of the drug (275 mg in total). In another case, 62-year-old man of gastric cancer accompanying carcinomatous peritonitis, after the repeated treatments with the drug (1 intraperitoneal injection, 50 mg; 12 intravenous injec-

tions, 25 mg each), tumorous ascitic fluid diminished considerably. Any unfavorable side effects were not observed except slight leucopenia and thrombocytopenia.

附 議

勝沼精蔵：Nitrogenmustard の中から石館教授が撰出したものの中 (X) (Y) (XY) のものを疑はれ私は Y が Z よりも臨床的には副作用がなく使用できやすいものではないかとの返答をしたのであるが、Y の構造があとでどうも明らかでなかったので、ただ今の Nitromin が一般に提示されて、今日にいたったが私はあきらめられず十年をまつた。石館、桜井、田坂氏等は私の臨床薬理の所見を信じて今日いま報告のような私の所見を思わせるようなものが得られたとの報告を今ききました——この先どうなるか不明ですがこの近所の諸君から一言を強いられたのでいきさつを一す。皆様の十年間ねばってくださったことを学究の立場から感謝し私の臨床薬理学的研究の一つの仕事となったことを喜びます。ただこれが Y そのものであったかどうかはなお将来に問題をのこしていると思います。

70. STUDIES ON THE CYTOSTATIC ACTIVITIES OF ENDOXAN, TRENIMON, AND E 39

TOSHIAKI EBINA, MASAHIRO SATO, KAZUO SATO,
MINRO WATANABE and NOBUKO OKAMURA

(Research Institute for Tuberculosis and Leprosy, Tohoku University)

Cytostatic activities of new alkylating agents, Endoxan, Trenimon, and E 39-soluble, against Yoshida sarcoma and the solid and ascitic tumor of Ehrlich carcinoma were examined together with cytological observations.

In the case of Yoshida sarcoma, considerable prolongation of survival time occurred by intraperitoneal injections of each agent (approximately $1/4$ LD₅₀ per day) for five days beginning 24 hours after inoculation. A single intraperitoneal injection of Endoxan ($1/2$ LD₅₀) was also effective. In the case of Ehrlich carcinoma, however, no significant effect was observed in the case of Endoxan or E 39. Prolongation of survival time was observed only when Trenimon was injected intraperitoneally for five days from 24 hours after inoculation. Marked decrease of packed-cell volume was also observed on animals treated with Trenimon.

Pronounced inhibitory effects upon the solid Ehrlich carcinoma was also attained by the intraperitoneal injections of Endoxan or Trenimon performed daily for one week beginning not only 24 hours but also one week after the inoculation. Inhibitory effect of Endoxan was so strong that the tumor could hardly be detected in almost all animals, and effect of Trenimon was as strong as that of Nitromin. E 39 showed no effect upon the solid tumor.

Cytological effects were examined at certain times after the single intraperitoneal injection of each agent ($1/4$ LD₅₀) which was performed on the fourth day of intraperitoneal inoculation of Yoshida sarcoma cells. The changes observed with Endoxan

or Trenimon were very similar to those produced by Nitromin; considerable number of abnormal mitosis became observable after 24 hours and these figures were more pronounced after 48 hours. Scattering, adhesion, granulation, or bridge-formation of chromosomes were observed in these cells. And the cells were swollen in general. Number of PAS-positive cells also increased 48 hours after the injection of Endoxan.

Electronmicroscopic observations revealed increased electron density of cell nucleus in general, increasing the density inside along the nuclear membrane. Elongated figures of endoplasmic reticulum were seen in cytoplasm.

71. ANTITUMOR EFFECT OF ENDOXAN ON THE TRANSPLANTABLE AND SPONTANEOUS ANIMAL TUMORS

TETSUO MINESHITA, TOORU IWAKI, KENJI YAMAGUCHI
and HEIZO TSUJII (Shionogi Research Laboratory)

Bashford carcinoma 63, Crocker sarcoma 180, Ehrlich carcinoma, NF sarcoma, Walker carcinosarcoma 256, Yoshida sarcoma and mice leukemia SN 36 were used as the transplantable animal tumor, and four spontaneous mice adenocarcinoma originated from the mammary gland found in the dd-s homogenous strain of mice were employed as the spontaneous animal tumor to examine the antitumor effect of Endoxan. Further marketed antitumor drugs such as Nitromin, 6 MP, Thio-TEPA and Mitomycin C were used at the same time to compare the effect of each drug. The drug was evaluated according to both macroscopical and microscopical findings.

1. *In vitro* test (contact test): Endoxan showed little effect and Nitronin much effect by the same concentration.

2. *In vivo* test: Prevention of tumor development with intraperitoneal administration of doses of $LD_{50} \times 1/20$, $LD_{50} \times 1/10$, and $LD_{50} \times 1/5$, Endoxan showed the most prominent effect of all the drugs used. The tumor responding most favorably was the Yoshida sarcoma and the worst the Ehrlich carcinoma.

3. Curative test: Even greatly enlarged transplanted tumors were affected by Endoxan fairly well, while all other drugs showed only a poor effect.

A much interesting effect was seen in the curative test with Endoxan on four mice with spontaneous adenocarcinoma. The four tumors each showed a different response of the grade ranging from (-) to (++), though the animals were of homogenous strain and treated in the same manner with the same dose at the same time.

72. EXPERIMENTAL AND CLINICAL STUDIES ON ENDOXAN

JUNICHI TOKUOKA, SHIGERU MIZOTA, HIROSHI SATO,
TAKUMI TSUNEMATSU, HIDETARO TOKUYAMA,
HIROSHI SATO* and TAZUKO TASHIRO*

(Kyoundo Hospital, The Medical Institute of Sasaki Foundation;
The Iatrochemical Institute of Pharmacological Research Foundation*)

(1). The effect of Endoxan upon 16 ascites tumors of the rat was studied. The examination was performed with special respects to the damage of tumor cells as well as the prolongation of survival time of tumor bearing animals induced by Endoxan treatment. Remarkable inhibitory effects on the Yoshida sarcoma and the ascites hepatomas, AH 13, 66F, 99, 130 and 322, were demonstrated.

(2). Comparative studies on Endoxan and Nitromin were carried out. No difference was found in the maximal tolerated dosis as well as the minimal effective dosis of the two drugs in the rat and Yoshida sarcoma. However, the maximal tolerated dosis of Endoxan in the dog is larger as 5 times as that of Nitromin. That a larger animal can tolerate a large amount of Endoxan may suggest the less toxicity of the drug in a larger animal as man. The duration that Endoxan remains effective on the Yoshida sarcoma cells in the abdominal cavity of rats after the intraperitoneal injection is 3 hours, while that of Nitromin is 90 minutes.

(3). 75 cancer patients were treated with Endoxan. 38 cases out of them were treated by combinations of the surgical operation and the Endoxan injection. The remaining 37 patients received only Endoxan treatments. The drug used per diem is 100 mg and about 1,000 mg in total were used in each case. In 14 of all the cases, remarkable inhibitory effects such as reduction of tumors in size as well as diminution of tumorous ascitic fluid were found. Subjective effects were also observed in 37 cases. These are improvement of appetite, disappearance of discomfort, etc. Side effects observed, such as leucopenia and thrombocytopenia, were very slight. It was demonstrated that the intraperitoneal injection of the drug caused the least side effect among various routes of applications tested.

(厚生省研究費による)

73. CLINICAL EXPERIENCE WITH ENDOXAN

YOSHINAGA TANAKADATE, ISAJI OHASHI and HIROSHI NAKASEKO

(Department of Internal Medicine, Nagoya National Hospital)

Endoxan was used for 25 cases with various malignant tumors. One hundred mgm. of the agent dissolved in 20% glucose solution was intravenously administered once a day. The total amounts used were varied between 600 and 4500 mgm. The subjects included were 7 cases with primary and 3 cases with metastatic lung cancers; 3 cases with hepatoma; one with laryngeal cancer; one, maxillary cancer; one, ovarian cancer; one, cancer of the colon; one, chordoma; one, neurofibromatosis Recklinghauseni; one, tumor of the spinal cord; 3, acute leukemia; 2, chronic leukemia.

Following the administration of Endoxan, recession of pleural effusion and edema on the thoracic wall was observed in 4 cases with lung cancer. In the case with chordoma, swelling of lymphnodes as well as splenomegaly diminished. In only one of the acute and one of the chronic leukemia cases, improvement in the blood picture was noted.

In none of the cases with lung cancer, improvement in roentgenographic findings was experienced. In general, there were no cases in which marked improvement was obtained.

Side effects were seen in 15 cases (60%) of 25 cases. In the side effects were included 4 anorexia, 2 nausea and vomiting, 1 vertigo, 1 feeling of fever, 1 diarrhea.

Slight leukopenia was noted in 18 of the 20 cases. In none of these cases it was of such a degree as required discontinuance of the agent.

74. CLINICAL TRIALS OF ENDOXAN, A NEW ANTITUMOR AGENT

YAEMON SHIRAHARA, YOSHIO NAKAMURA, KATSUJI SAKAI

and KENICHI HASHIMA

(Department of Surgery, Medical School, Osaka City University)

We are clinically investigating the effects and the side-effects of Endoxan, an antitumor drug. The drug is administered intravenously or by intubation into arteries, in the dosage of 100 to 200 mgm per day.

Up to now, 6 patients out of 32 have demonstrated clinical improvements such as decrease in size of the tumor, of the 'cancer milk' and of amount of ascites.

Complete blood counts on the peripheral blood checked every 5 days during the period of the administration, showed leukopenic tendency in 10 patients among 32. However, the rest of 32 cases hardly revealed leukopenia. In 4 cases, on the contrary, who received more than 1 gm of Endoxan in the total dosage, leukocytosis was observed.

No significant abnormality was detected in the patients regarding the hepatic functions, electrolytes, platelet count and prothrombin time. No untoward side effects were noticed, too.

75. STUDIES ON CHEMOTHERAPY OF MALIGNANT NEOPLASMA (VIII)

YUZO TAKEMASA, TADASHI SUGIYAMA, TADASHI KIMURA,
and YOSHIYUKI KOYAMA
(1st National Hospital of Tokyo)

1) Endoxan was administered intravenously to 47 patients suffering from malignant diseases at a level of 100 mg/day and occasionally it was injected locally, and given orally. The total dose was 0.4 g to 9.0 g. Clinical improvement, namely regression of the tumor, was seen in nine out of 47 patients. These nine patients included cases with reticulosarcoma, two leucemias, three carcinomatous peritonitis, lung carcinoma, and paratoid carcinoma. Objective improvement was seen in the cases treated with more than 3 g of Endoxan. Side effects were noted as follows; anorexia 3 cases, nausea 1, leucopenia 11 and thrombopenia 5. The clinical effect of Endoxan was just the same as for the other alkylating agents, and the side effects were observed less than with TSPA.

2) E 39 was administered to 14 patients with malignant diseases, intravenously at a level of 10 mg/day. The total dose was 0.07 g to 0.5 g. Clinical improvement was seen in 3 cases, lung carcinoma, lung metastasis of Grawitz tumor, and reticulosarcoma. Side effects were as follows; anorexia 1 case, nausea 2, fever 2, exanthema 2, leucopenia 3 and thrombopenia 5.

3) Nitromin D was administered to 19 patients with malignant diseases. Clinical improvement was seen in 4 cases: two reticulosarcomas, Hodgkin's disease, and carcinomatous peritonitis. Side effects were as follows; anorexia 5 cases, nausea 3, vomiting 1, leucopenia 6 and thrombopenia 4. The clinical effect of Nitromin D was just the same as NMO and the side effects of the former were less than the latter.

76. FUNDAMENTAL AND CLINICAL STUDIES ON MH

NOBORU IJIMA, KIYOSHI MATSUURA, AKIRA UENO,
KICHISHIRO FUJITA, TATSUO AIBA, HITOYA UKISHIMA,
FUJIO NOHARA and TAMEHISA KOSHIZUKA

(Department of Surgery, School of Medicine, Tokyo University)

Experiments have been made on the effect of MH in the relation between tolerance of an individual to general aggression and his antitumor action. When the resistance to an aggression is increased, antitumor action is also increased. MH was found to have such an effect. Furthermore, as the resistance is continuously raised, finally it becomes exhausted and anti-tumor action also disappears. Then, it was found that it resulted even in accelerating the growth of a tumor.

From this point of view, continuous administration of MH for patients with malignant tumors may be rather harmful while intermittent administration would be most effective.

77. DISTRIBUTION OF ^{203}Hg -HEMATOPORPHYRIN- Na_2 IN TUMOR BEARING MICE AND CO-ACTION OF X-RAY AND HEMATOPORPHYRIN- Na_2

HIROAKIRA SOEDA and SHIGEAKI OKAMURA

(Department of Radiology, Faculty of Medicine, Kyushu Univ.)

It was reported by one of the authors that the effectiveness of the combined administration of Hg-Hematoporphyrin- Na_2 (MH) and x-ray irradiation on the Ehrlich-ascites solid tumor of mice is larger than that of single administration of any one of these two treatments. To explain this result the distribution of MH in mice, and the influence of MH, and Hematoporphyrin- Na_2 (HP) on the lethal effects of the total body irradiation of mice were investigated.

The results of the present studies are summarized as follows:

1) Distribution of intraperitoneally injected ^{203}Hg -Hematoporphyrin (R-MH) in tumor bearing mice and normal mice were checked every five days after injection by γ counting of organs listed below. Distribution of R-MH are as follows; in kidney 32.9~78.6, liver 17.6~34.8, tumor 6.9~17.4, lymph node 2.6~10.1, spleen 2.7~7.7, lung 2.7~5.9 and blood 1.0 (concentration of R-MH in blood is taken as 1.0).

2) 750 r of total body x-irradiation was performed 24 hours after intraperitoneal injection of $1/2$ LD₅₀ of MH. Survival curve of MH injected group was compared with only irradiation group, and slight shortening of survival time was observed in MH injected group up to 9 days after irradiation.

3) 9.17 mg of HP is equivalent to $1/2$ LD₅₀ (12.0 mg/kg) of MH, but HP is less toxic compared with MH, as its molecule does not contain Hg atom. Therefore, 9.17 mg/kg and 5×9.17 mg/kg of HP were intraperitoneally injected and 550 r of total body x-irradiation was added 24 hours after injection. As control, only H.P. injection (5×9.17 mg/kg) groups and only irradiation groups were prepared. Combined administration of HP and x-irradiation is more effective than that of only irradiation, although HP injection alone does not cause any lethality in mice even at high dose of 5×9.17 mg/kg. Survival rates at 30 days after administration were; HP (5×9.17) +550 r, 76%; only irradiation (550 r) 88%, only HP (5×9.17 mg/kg) 100%.

78. STUDIES ON CHROMOMYCIN, EXPERIMENTAL AND CLINICAL, AND ADENOSINE-DEAMINASE ACTIVITY OF SERUM OF CANCER PATIENT

SADATAKA TASAKA, KEIMEI MASHIMO, YOSHIO KURODA,
TOSHIO HARADA, KIHACHIRO SHIMIZU, MASAMI HATAKEYAMA,
OTOHIKO KUNII, EIACHIRO YAMADA, TSUNEO JINDATE
and KAORU SHIMADA

(Department of Internal Medicine, Faculty of Medicine, University of Tokyo)

I. Experimental studies on mechanism of action of Chromomycin.

1. *In vitro* experiments: The antibiotic activity against *B. subtilis* of chromomycin was not found to be lowered by addition of serum or erythrocyte of rabbit. On the other hand there was an apparent reduction of its activity when incubated at 37°C for 30-60 min. with emulsions of lung, spleen and kidney. It might be probable that Chromomycin was adsorbed or inactivated by them.

2. *In vivo* experiments: a) The concentration of Chromomycin in blood after its intravenous injection (0.5 mg/kg) fell down quickly and it was excreted in urine about $1/4 \sim 1/5$ total dose injected within 5 hours in rabbit. b) The distribution of Chromomycin *in vivo* has been investigated with emulsions of several organs of rabbit after its intravenous injection. It has been shown that Chromomycin had an affinity, more or less, to the lung, liver, kidney and specially spleen.

II. Clinical studies on Chromomycin.

Among 22 patients of various malignant diseases treated with Chromomycin, ob-

jective effects have been seen in 6 cases. In 3 cases with peritonitis carcinomatosa, ascites was decreased. In one case of postoperative cutaneous metastasis originated by gastric sarcoma, a shrinkage of its size, but only temporarily, and in some other cases remissions of jaundice due to gastric cancer or bloody sputa following lung cancer were observed.

It was characteristic for Chromomycin that leucopenias as the most popular side effect were quite rare. However when intravenously injected, its subcutaneous leakage easily suffers from vesicle or necrosis in its local site. Gastrointestinal complaints and influences on liver or renal functions were scarcely observed.

III. Adenosine-deaminase activity of cancer patient serum has been reported to be increased by Letnansky et al. in 1958.

In healthy 38 cases its normal level was found to be calculated by us as 0.284 unit (1 unit: the activity deaminating $1\mu\text{M}$ adenosine per ml serum). On the other hand cancer patients showed in general a tendency to increase in its activity. In particular, in case of acute leukemia, malignant lymphoma or peritonitis carcinomatosa a marked increase was shown, their averages were 0.824, 0.815 and 0.654 respectively. In other cases of malignant diseases its activity was variable in each case and not so few showed its normal level. In non-malignant diseases, the levels did not differ so much from normal but in acute inflammation sometimes its activity was shown to be elevated. In some cases of leukemia or malignant lymphoma, there was relative correlation between their clinical courses and the activity of adenosine-deaminase.

79. CHEMOTHERAPY IN CANCER PATIENTS TREATED WITH MITOMYCIN-C AND BONE MARROW TRANSPLANTATION

TAKAO HATTORI, GENSCHIRO FUJII, KAZUTAKA ASHIKAWA,
KIKUO MOTOYA and YUKIO ISHIBASHI

(Department of Surgery, Institute for Infectious Diseases, University of Tokyo)

This report concerns the clinical experience in the use of Mitomycin-C followed by the transfusion of the autogenous or homologous bone marrow in 19 incurable cancer patients during the past one year. In the first group (10 cases), 50-192 mg of Mitomycin-C (MC) was injected intravenously within one or a few days. The damage of the bone marrow was repaired well by the bone marrow transfusion, but the other side effects such as hemorrhagic diarrhea, anorexia, stomatitis and depilation were observed in various grades. Although clinical improvement was seen in nine of the 10 patients, three of them died from bacterial infection within two weeks, three died of the same cause within four weeks and the others died from the relapse

of the tumor after three months. The autopsy revealed that the extreme atrophy of the lymph-systems and the multiple ulcers of the large intestine were responsible for the fatal outcome.

In the second group (6 cases), 10 mg of MC was injected intermittently once a week about 60 mg totally with the following transfusion of the bone marrow. The side effects in this group were observed very slightly. Excellent improvement was seen in two cases, but no improvement also in two. It appeared that the result of this second treatment was entirely dependant on the sensitivity of the tumor cells to MC.

In the third group (3 cases), 20-30 mg of MC, as a single dose, was injected under the interruption of cranial mesenteric and splenic arteries for 30 minutes in order to keep the lymph-systems and the intestinal mucosa from being exposed to the high concentration of MC in the circulating blood. Excellent improvement was seen in one patient with lung cancer, who survived for four months after the administration of MC without initial complaints.

80. STUDIES ON THE INTERMITTENT APPLICATION OF MITOMYCIN C

ISAMU USUBUCHI, SHOICHI OBOSHI, JUNNOSUKE YOSHIDA,
MICHİYOSHI SUGAWARA, and TOSHIRO HONGO

(Department of Pathology, School of Medicine, Hirosaki University)

Hybrid rats transplanted intraperitoneally with Yoshida sarcoma, diploid Hirosaki sarcoma, Usubuchi sarcoma, hepatoma 130 or hepatoma 7974 respectively were treated by the intraperitoneal administration of Mitomycin C of 2 mg/kg or 1 mg/kg or 0.5 mg/kg 48 hours after the transplantation. Some of the rats received the second and the third injection in the same manner respectively after the interval of 1 week. Except for the cases of hepatoma 7974, most animals were cured of tumor by the single injection of 2 mg/kg or 1 mg/kg.

In half of the experimental cases, including the cases of hepatoma 7974, tumor cells disappeared from the ascites and, thereafter, nodular tumors in the peritoneal cavity diminished gradually by the single intraperitoneal administration of Mitomycin C of 2 mg/kg or 1 mg/kg which was carried out 5~7 days after the intraperitoneal transplantation.

The data seem to show that the tumor cell destroyed by the single injection of the drug produce antibodies, which may act on the remaining tumor cells. The

same mechanism can be expected in the treatment of a human generated tumor.

(文部省科学研究費による)

附 議

秦 藤樹: Mitomycin を間歇投与すると効果があるという御報告に対し追加します。

Ehrlich 癌 (皮下腫瘍) を移植した mice に Mitomycin の投与全量を同一にして毎日連続分割投与群、3 日間隔の投与群、1 週間間隔投与群の 3 群に分ち、制癌効果を比較検討したところ効果は 1 週間間隔の投与群が最も顕著であり次で 3 日間隔のものが続き連続投与群の効果が最も劣っていた。さらに制癌効果のみならず副作用の軽減が間歇投与群においてとくに 1 週間間隔の群において著明となった。

以上のことは Mitomycin のみならず Carzinophilin の場合さらにわれわれが最近発見した S-339 物質の場合においても同様の傾向が見られた。

次に併用実験として Mitomycin と Carzinophilin を combination して以上の実験を行つたがやはり間歇法がすぐれており、Mitomycin あるいは Carzinophilin 単独の場合よりも両者の combination の場合の間歇投与法が最もすぐれており一定量の組合せでは腫瘍の完全消失がその群の大部分のものに見られた。

以上は動物実験であるがこれを臨床的に確認していただきたい (なお成績の詳細は本年の国際癌学会シンポジウムで報告した)。

武田勝男: 日本における化学療法の研究に雑種動物が使われているが純系動物にその系統に発生した腫瘍を用いないかぎりいろいろな意味の免疫が発生する、これは注意しなければならない。

河村謙二: 腫瘍細胞が抗癌剤の大量一時的投与によって破壊されることによる免疫現象は次の腫瘍発育に抑制的に働くのではないかということは当然考えられるが、薬剤によって得られる抗腫瘍性耐性がどうして起るかということを含めてきわめて複雑な原因によるものと考える。単に免疫だけでは説明が困難なのではないか、私に次のような実験がある。抗癌剤を投与後採取した血液を移植後 6 日目の担吉田肉腫ラッテから採った腹水と混合して 37°C、30 分間 incubate したものを健康ラッテに移植するとこの腫瘍の発育はなんら影響をうけず単なる移植の場合と同様であるのに、このラッテ腹水を次の健康ラッテに移植するとこの二代目ラッテでは腫瘍の発育はいずれも抑制されその生存期間は著しく増大する。この次同様にして三代目を検するとこれはまったく初代と同様となる。すなわち二代目でどうして延命されているか、これは未だ私には充分解釈できぬが免疫その他耐性の出現と関係ある興味ある問題である。

石橋幸雄: われわれも癌の化学療法の効果の中に生体の免疫反応が関係していると考えている。その意味で演者の研究は興味深く拝聴した。10% の動物を殺す腹水型の腫瘍を皮下に移すとなかなか死ななくなる。Origin から離れた腫瘍に対して生体は抵抗を示すと解される。化学療法の実験において、皮下型、腹水型を持つ動物の方が、腹水型単独群よりも延命率が高い。その根底に免疫機序が関与していると思われる。臨床例においても、皮下転移を持つ患者は一般に薬剤に sensitive であるという事実が経験される。近藤達平: ただ今の御報告には臓器内転移についてはしらべておられないようであるが、われわれは 1956 年以来制癌剤使用による臓器内転移(増癌作用)について不十分な制癌剤の使用は非常に危険であることを主張してきた。1 カ月間隔に副作用の強い宿主抵抗を下げる制癌剤を臨床的に使うことは危険であると考える。

白淵: 私どもの実験では Mitomycin 使用の際に連日投与の場合より間歇投与または 1 回投与の方が副作用も少く、再発も少なかったことをみています。

81. STUDIES ON THE PREOPERATIVE USE OF ANTI-CARCINO- GENIC AGENTS (III): ADEQUATE TIME FOR ADMINI- STRATION OF ANTI-CARCINOGENIC AGENTS

KUNIO OKAJIMA, NOBURU SAKAKIBARA and KAZUO MORISHITA

(1st Department of Surgery, Medical School, Okayama University)

To study the adequate time for the concurrent use of anticarcinogenic agents at radical operation, experiments have been conducted with Nitromin using ascitic hepatoma MH 134 mice and C_3H xdd: F_1 mice, that proved to be 100 per cent certain of subcutaneous transplantation and ultimately die of tumor because of post-operative metastasis even after removal of nodular tumor by operation. Radical operation is performed 7 days after tumor transplantation for group I and 5 γ /g./day Nitromin is given for 8 consecutive days after operation. Group II are operated on 7 days after the transplantation and Nitromin is given in the same manner from the day after operation. For group III, 8 γ /g./day Nitromin is administered for 5 days from the fifth day after transplantation and operation is performed a day after the last administration. The same dosage per day is given to group IV for 5 days from the twelfth day after transplantation and is operated on the day after the last administration. The survival rates due to these treatments proved to be 57.1 per cent in group I, and 47.1 per cent in group IV, showing that the pre-operative administration is more effective than the post-operative one.

Next, in order to determine whether or not tumor cells circulating in blood can be a recurrent factor, the blood aspirated from the heart of cancer bearing animals was injected into the peritoneal cavity of normal mice and the manner of ascites formation in the group administered with Nitromin and in another group without drug administration was observed. As the result the rates of tumors metastasis proved to be 33 per cent in the group given the drug and 80 per cent in the group without drug administration. From these findings it can be said that even in the presence of tumor, anti-carcinogenic effect on tumor cells circulating in blood can be expected by administration of such drugs.

(文部省科学研究費による)

追 加 徳山英太郎, 徳岡淳一, 溝田 成, 佐藤 博, 常松 匠

われわれは今春の第3回癌シンポジウムにおいて, 共同研究者山田と共に, 癌細胞の結合性と転移と題して, 制癌剤を使用すると, 癌細胞の発育を抑えると共に, その結合性をますことを発表した。スライドのごとく AH 601 5日目の腫瘍を Tween 80 で解島するさい, 腫瘍動物にあらかじめ制癌剤を与えておくと, 解島が困難となってくる。この現象は制癌剤により発育を抑制すると, 腫瘍の剥離性が抑えられ

るという現象と考えられる。

また昨年9月日本消化器病学会で述べたが、胃内に移植した吉田肉腫腫瘍に対して、ナイトロミンを使用すると、決して胃腫瘍を胃内腔に剥離させるに十の作用はなく、むしろ一の作用があることを発表した。さらに今回の癌学会でも、血中癌細胞に対する影響を示した。

これに関連して現在次のような実験を行なっている。すなわち吉田肉腫を1000万個呑竜系ラットの右後肢筋肉内に注入し、これを外科的5日目に切除する。この際切除前にナイトロミンを1.0 mg 宛2日間使用しておくだけで、スライドのごとく単純外科的切除群に比し著明な生命延長を認める。制癌剤の術前使用を臨床例に行なう際に、直ちにこの事実をあてはめることはもちろんできないが、ある種の人癌では、このような効果を期待することも可能であろう。

すでに Stöger は 1953 年に Colchicin-Schütz なる言葉を提唱してをり、彼は術前2時間に 1.5 mg の Colchicin を筋肉内に注射している。私共もすでに臨床例において術前2日前より Endoxan を1日100 mg 2日間投与している。

附 議

今永 一：外科手術と制癌剤の併用という問題でもっとも重要な点は制癌剤の投与時期であると考え。それは腫瘍の移植前に制癌剤を投与すると移植癌の非常に悪化することのあるを認めている。したがって術前に投与する場合は、その投与量によってはかえって悪結果を来す場合があるのではないかと考えている。この問題は非常に大切なことと考えているので今後さらに検討を加えてみたいと思っている。

陣内伝之助：ただ今のお話のごときのことは東大清水外科教室の実験でも、術前投与の方がかえって悪い結果になっておりますが、これは健康な動物に制癌剤を投与した後に腫瘍細胞を移植しておられるからだと思ひます。わたくしの教室の実験は担癌動物に対して化学療法と手術を行なっておりますので制癌剤の投与が担癌動物であるのと正常動物であるのとで、大いに成績が異なることを私どもも血中 Properdin 値を示標として確かめております。先ほど臼淵教授の話されたように担癌動物に制癌剤を注射した場合には免疫ができるということも考えられると思ひます。

徳山英太郎：術前に制癌剤を使用すると、かえって手術後の転移を助長するのではないかと質問に対して、術前わずか48時間から少量を与えて十分な効果があるので、この程度のものでは個体の抵抗性を低下するような心配はないと思う。

武田：武田肉腫を2ヶ所に植え1側をとると他側は急激に増殖する、1カ所に植えて一定期間剔出すると内臓の転移が急激にふえる。この手術の際血液に腫瘍を移入しても影響がない、したがって手術による主腫瘍の剔出はなんらかの意味で他腫瘍あるいはすでにある散布果の増悪を結果する。以上の意味で手術前の化学治療がのぞましい。

82. STUDIES ON LOCAL ADMINISTRATION OF ANTI-CANCER AGENTS BY MEANS OF TISSUE CULTURE AND ELECTRONMICROSCOPY

KENJI KAWAMURA, SHIRO KITAGAWA, KOJI NISHIMURA,
SHOJI KAWAMURA, AKIRA OOMOTO, TORU SATO,
MASAHARU NAKAJIMA, TAKAFUMI EBISE and RYOHEI OKAMOTO
(2nd Department of Surgery, Kyoto Prefectural University of Medicine)

It has been recognized to be very important to apply chemotherapy to surgery, but the determination of effective concentration in blood and sensitivity of the anti-cancer agents is still very difficult problem.

The authors studied the influences of the concentration of several sorts of agents on the total growth-coefficient, using the extracorporeal tissue culture (heart and spleen) of the chicken embryo.

RC-4: The proliferous depression begins from 32/ml for heart and 0.01/ml for spleen, and becomes very remarkable in 100/ml.

Nitromin. Tespamin, Carzinophilin; The depression appears in about 10/ml.

Merphyrin and Azan: The appearance of depression needs more than 200/ml.

The finest cell structure of the proliferated tissue cultured with 10/ml anti-cancer agent was investigated under the electron-microscope, and then no notable changes except smaller mitochondria in size was recognized if added with Carzinophilin.

The authors made a trial to use tissue culture solution added with the venous blood adopted from the portal vein from 2 seconds to 2 hours after Tespamin was infused into gastric artery and femoral artery of the experimental dogs. So the growth depression is stronger in 1 to 5 minutes solutions, and decreases in later solutions, but is still kept in 3 hours solution.

Also when adopted from portal vein and femoral artery after infused into gastric artery, no difference in the depression effect for both the former and the latter is recognized if the infusion pressure remains within 200 mmHg, but if over 400 mm Hg the depression effect of the portal vein blood is kept strong after 5 minutes.

However when the anti-cancer agent was administered through vein and artery to the hand-cancer caused by the radio-active ray, possibly proliferable cancer cells in the improved tissue were seen under the electron-microscope.

Adenine and pan-amino-acids solution which is said to be able to antagonize the side-effects of the anti-cancer agents, decrease the growth depression effect of Tespamin in the tissue culture, but are not seen to accerelate the proliferation than usual.

The therapeutic effect for the rat transplanted with Walker's carcinosarcoma was observed to be best in the case used Nitromin plus Adenine among the three groups, Nitromin, Adenine and Nitromin plus Adenine.

83. EXPERIMENTAL STUDIES ON LOCAL APPLICATION OF ANTICANCEROUS CHEMOTHERAPEUTICS IN SURGICAL OPERATION (II)

CHUZO OTA, MITSUE MIURA, YUKINAO HARIMA
and YUKIO TAKEDA

(Surgical Clinic, Faculty of Medicine, Hirosaki University)

1) The ratio of increase of *E. coli* was examined *in vitro* by using anticancerous agents (Nitromin, Tespamin, Carzinophilin, Mitomycin C and Sanamycin) in combination with antibiotics (Streptomycin, Terramycin, Chloromycetin etc.).

2) The effect, either synergic or antagonistic, was depended upon various combinations of antibiotics and anticancerous agents and these concentration. When the low concentrated anticancerous agents were used jointly with antibiotics the effect against the bacterial growth was weakened as compared with that of antibiotics alone.

3) We concluded that the most favorable combination for the local application in operation of cancer are as follows; Mitomycin C and streptomycin or Terramycin, Tespamin and Streptomycin, Nitromin and Achromycin, Carzinophilin and Chloromycin, Carzinophilin and Chloromycetin etc.

4) In our previous report we described that Tespamin was a favorable agent for wound healing. So for the local application, the use of Tespamin in combination with Streptomycin may be a method of choice.

84. EXPERIMENTAL STUDIES OF ANTICANCER AGENTS COMBINED WITH SURGERY

RYUJI ONO, MASANOBU AKAGI, RYOYA TOMOJIRI,
HISASHI SAITO, KUNITOSHI KITANO and FUMIO KATSUHIKA

(2nd Dept. of Surg., School of Medicine, Kumamoto Univ.)

It would be preferable to use anticancer agents together with surgical treatment of cancer to protect its recurrence.

The fundamental experiments for anticancer agents especially in surgical field were

planned, because there were very few studies reported.

All dogs have had gastrectomy (Billroth-II) and several kinds of anticancer agents were used for 10 days before and after operations.

Clinically liver funktion, serum protein and hemogram including bone marrow examination were observed. Histologically area of gastro-jejunosomy, of entero-entero anastomosis, skin wound, liver, spleen and kidney etc. were studied.

The chief disturbance was found in hematologic studies clinically and there were slight changes in liver funktion tests and serum protein.

No big disturbances were found for intestinal anastomosis pathologically in all cases, but reepithelisations in the area of intestinal anastomosis were probably less in the groups used anticancer agents compared with the control group.

Several grades of degeneration in the liver and kidney and marked atrophy of spleen were observed especially in the groups which were used anticancer agents.

We consists strongly there were no arguments to use anti-cancer agents by surgical treatment of cancer with its suitable management.

追 加

三浦光恵：外科手術にあたって、制癌剤が手術創の創治癒に対して、いかなる影響を与えるかに関し追加する。1) 雑種白鼠で切開創を 5~7 日目に張力を加えて、けん引し離開に要する重量 (gr) を求めると、Thio-TEPA が最も張力を必要とし、Nitromin 群は簡単に離開する。これらのことは組織学的にも証明された。2) 臨床例について、Thio-TEPA 10 例、Nitromin 3 例について、後者 3 例に組織の壊死が認められた。動物実験の成績と一致することをスライドで示した。

85. CHEMOTHERAPY OF INTRACRANIAL TUMORS

ISAO OSHIRO, SATORU KUSAMA, TAKAYASU YOSHIDA,
TAKASHI IWASAKI and KENJI MATSUOKA

(1st Dept. of Surg., Tokyo Univ.)

The effect of the cancer chemotherapy with or without operation on cases with intracranial tumors who had been admitted in our clinic was investigated symptomatologically and the follow-up studies were made. The 5-year survival rate of 1254 cases of intracranial tumors was 48%.

Of these, 116 cases were treated with Nitromin, Thio-TEPA or other anticancer drugs before and/or after operation. The follow-up studies revealed that the chemotherapy had no life-prolongation effect in all types of intracranial tumors, except meningosarcomas and intracranial cancer metastases where some beneficial effects were seen. The improvement of the symptoms was obtained in only 13 cases.

A single dose of ^{32}P -labelled Thio-TEPA (containing about $100\mu\text{c } ^{32}\text{P}$) was given to adult dogs into the lateral ventricle or into the common carotid artery, and the

concentration of the drug in the cerebrospinal fluid, the brain tissue and the blood were estimated by means of GM counter. The cerebrospinal fluid was obtained from the great cistern and the brain tissue from the praecentral area and the blood from the femoral artery.

By the intraventricular application, the concentration of the drug in C.S.F. was highest about one hour after administration and gradually lowered subsequently, whereas the concentration level in the brain tissue was only about one fifth of that in C.S.F. at the highest and almost constant from one to six hours after administration.

By the intracarotid application, the concentration level of the drug in the brain tissue and C.S.F. were almost the same curves and showed similar to that in the brain tissue by the intraventricular administration.

Since no difference of the concentration of the drug in the brain tissue was found between the intraventricular and the intracarotid administration, with regard to Thio-TEPA, no different effect can be expected from these two ways of application.

(文部省科学研究費による)

86. THE STUDY ON THE CHEMOTHERAPY OF GASTRIC CANCER, PRELIMINARY REPORT

TATSUZO OHARA

(Cooperative Studies of National Hospitals in Japan, on Cancer Chemotherapy,
Committee of Gastric Cancer)

Here presents the report of the cooperative study on chemotherapy against cancer of the stomach. The study was made by nine National Hospitals in Japan (SENDAI, CHIBA, TOKYO-1st, TOKYO-2nd, NAGOYA, KANAZAWA, KYOTO, OSAKA and TSUKUSHI).

The purpose of this work is to ascertain the effects of anticancer chemotherapy on patients who had radical operations for stomach cancer. These patients were divided into 3 groups by the random-sampling method. The 1st group was given Tespamin, the second, Mitomycin, while the last was the control group.

The dosage of anti-cancer drugs were as follows: Tespamin, 0.2 mg/kg daily, started from the day of the operation, until the total dose was above 50 mg; Mitomycin, 0.08 mg/kg daily and continued in the same manner until it was over 40 mg.

To avoid mechanical spread of cancer cells, 5 mg of Tespamin, or 4 mg of Mitomycin was sprayed into the peritoneal cavity during the operative procedure.

For statistical purposes, patients were divided into 9 subgroups, by age and by

locality of lesion.

By the end of October 1960, 292 cases were followed up. Of 66 cases, in which over 1 year had elapsed from the beginning of chemotherapy, the number of recurrent cases (including fatal cases) were: Tespamin-group 8/26, Mitomycin-group 9/23 and control 10/17.

Although it is not conclusive, the results of this study indicate that patients receiving chemotherapy with radical gastrectomy, have a better prognosis than those of the control group.

附 議

今永 一： 癌の根治手術成績の向上に対して制癌剤の使用はどの程度の効果があるかの検討は非常に大切なことであるが、その判定は多数の因子が関与するので非常に困難である。欧米の癌研究者もこの点についてはいろいろ検討を加えているようである。先日アメリカの Rosewell Park Memorial Institute より発表された成績では制癌剤の効果は認められなかったと述べている。この問題は多数の症例について多数の経験者の検討によって解決さるべきものと考えている。

87. CHEMICAL EXPERIENCE OF ANTICANCER AGENTS IN GYNAECOLOGICAL FIELDS

MINORU ISHIHARA, YOSHIRO KAWASHIMA, TADASHI UCHIDA,
SHIGEHICO INO, TSUTOMU NAKATSUKA, KAZUKO KATO
and MITSURU MIKI

(Department of Obstetrics and Gynaecology, School of Medicine, Nagoya University)

We report here the results after we used various anticancer agents such as 8-Azaguanine, Mitomycin C, Chromomycin, Endoxan, B 3231 etc. upon 400 cases of sexual cancers (uterine cancer, ovarian cancer, malignant chorionepithelioma). As for the methods, intravenous and local injections were used for inoperable patients. Operable patients were given 10 mg for 3 days before the operation and 2 mg for 10 days after it. Among 47 cases given Mitomycin C, 30 cases (64%) got better. Though, leucopenia and haemorrhagic tendency as side effects should be noticed. The former happened to reach nearly down to 1000 when 50~60 mg were given, and the tendency strengthened with radiation therapy. The latter seemed to base on the decrease of thrombocytes. Therefore to adopt precautionary measures it had better use leucocytosis-promoting substances and hemostatics. As hemostatics we used fresh blood-transfusion before the operation, we have faced with no inoperable cases even though the bleeding was more than usual. The histological figure improved in 6 cases among 8 cases (74%). In Chromomycin group 11 cases out of 17 cases (65%) recovered. We could use the agent for a long time because it accompanied little side effects. Besides in the histological figure it was effectual. Only in 6 cases Endoxan was used, and

the side effects such as leucopenia and loss of hair were observed. But we experienced 1 case in which malignant chorionepithelioma yielded metastasis to the lung recovered after 2600 mg of Endoxan had been given. B 3231 was used in 3 cases. When 1 mg was injected continually nausea and vomiting were hard, but 0.1 mg was injected such effects were not observed. At the end, blood catalase and β -glucuronidase were examined as the indicators of the decision of effects. In Mitomycin group, the former rising and the latter decreasing, we were able to guess the effectiveness, but in Chromomycin group they showed no change, and in Endoxan and B 3231 groups also they seemed not to have any effects.

88. ON THE FIVE YEAR SURVIVAL RATE IN THE CASES OF CANCER OF THE UTERINE CERVIX WHICH TREATED BY 8-AZAGUANINE AS ADDITIONAL TREATMENT AGENT

SEIICHI YAMAMOTO, YOSHIRO KAWASHIMA, TADASHI UCHIDA
and TERUO MASUKAWA

(Department of Obstetrics and Gynecology, School of Medicine, Nagoya University)

1) In our department 256 cases of uterine cervix cancer were treated for a period of 3 years and 3 months from January 1952 to March 1955. 231 cases including I-stage 15, II-stage 74, III-stage 93, IV-stage 49 out of 256 cases were treated with 8-azaguanine as an additional treatment agent, and its treatment effect was studied statistically on the basis of five year survival rate.

2) The five year survival rate of 115 cases treated by surgery and 8-azaguanine was 75.7%, and that of 116 cases treated by radiotherapy and 8-azaguanine was 32.7%. The five year survival rate of a total case was 54.1%.

3) The five year survival rate according to progressing stage was 93.3% in the first stage, 81.1% in the second stage, 38.7% in the third stage and 30.6% in the fourth stage.

4) When the above described results were comparatively studied, these results were superior to those that obtained by the treatment without using 8-azaguanine. Therefore, the usage of 8-azaguanine seemed to be significant in the treatment of uterine cervix cancer.

(文部省科学研究費による)

89. CLINICAL AND PATHOLOGICAL STUDIES ON CANCER OF URINARY BLADDER WITH ANTICANCER DRUGS

JUN'ICHI OMURA, KEN'ITSU OKITA and SUMIO TASAKA

(Department of Urology, Medical School, Okayama University)

Carzinophilin, Mitomycin C, and Thio-TEPA were administered to 32 cases of cancer of urinary bladder and cytological changes of the desquamated cells in urine and histopathological changes of tissue were studied. All of these cancers had their origin in transitional epithel, and almost all the patients were discovered with hematuria in early stage and diagnosed by cystoscopy.

Anticancer drugs were administered in local (local injection) or general, and the changes compared before and after administration. To 17 cases administered in general; Carzinophilin 7, Mitomycin C 5, Thio-TEPA 5, and 15 cases in local; Carzinophilin 7, Mitomycin C 6, Thio-TEPA 2.

1) After administration of 50000 u.-190000 u. of Carzinophilin in general observed the contraction of tumor 3 in 7 cases, degeneration of nuclei in the desquamated cells and slight proliferation of fiber in tissue.

2) After administration of 10000 u.-87000 u. of Carzinophilin in local observed the contraction of tumor 6 in 7 cases, and dissolution of nuclei or vacuolation of cytoplasm in the desquamated cells, cellular infiltration or necrosis in tissue.

3) After administration of 40 mg of Mitomycin C in general observed the contraction of tumor 2 in 5 cases and remarkably decreased stainability in the desquamated cells and falling of the tumor cells in local.

4) After administration of 2.4-20 mg of Mitomycin C in local observed the contraction of tumor 3 in 6 cases and degeneration of desquamated cells, infiltration or bleeding in local, but the reaction in local seemed milder than the cases of Carzinophilin.

5) After administration of 45-120 mg of Thio-TEPA in general observed the cystoscopic changes 2 in 5 cases and decrease of cytoplasm in the desquamated cells, atrophy or disappearance of cytoplasm and proliferation of fiber in tissue.

6) After administration of 90 mg of Thio-TEPA in local observed the contraction of tumor in all cases and pyknosis, fibrous proliferation.

In summary administration of anticancer drugs in general to cancer of urinary bladder which have their origin in transitional epithel was not so effective, while administration in local obtained some effective changes, and concerning our observations Carzinophilin was most effective, then Mitomycin, Thio-TEPA.

(文部省科学研究費による)

90. STUDY OF THE SERUM IRON LEVEL FOR EVALUATION OF ANTITUMOR AGENTS

TSUYOSI KUSUMOTO, JYUNICHI KOBAYASHI
and HIDEYUKI KAMEYAMA

(1st Departement of Surgery Medical School, Okayama University)

We confirmed that patients of gastric cancer could be prevented from the subjective and objective side reactions by preoperative administration of Mitomycin C by using blood transfusion or transfusion. Serum iron in gastric cancer decreased by the decrement of iron absorption from gastric and duodenal membrane by the growth of cancer and attendant gastritis, and by the bleeding from cancer and by toxohormone.

Under the conditions of the hemostasis by gastric resection and of the absorption of iron the definite residual stomach, supposing Mitomycin C is effective on cancer, serum iron should increase more than that of the control. And then, we divided 36 cases of gastric cancer into four groups, namely group of the control, group of preoperative administration of Mitomycin C, of postoperative administration, and pre-and postoperative administration, and studied on the efficacy of Mitomycin C.

We observed serum iron, and usual blood test on the entering day, on the day before operation and every five days after operation and the alleviation of symptoms day by day in each of groups.

There were remarkable changes in usual blood tests.

As regards serum iron, it reached its maximum in the control within 20 days, but decreased slightly in 25 days, and especially, in the 2 cases of test laparotomy, it decreased gradually from day to day, and then general conditions took a serious turn.

But in groups administered with Mitomycin C, serum iron increased more than the control including cases of test laparotomy, and the alleviation of the subjective and objective symptoms was better than the control. Next, we were assured that serum iron reflected alleviation or aggravation faithfully. Therefore, it is believed that these findings would prove to be a useful index for judging the effect of antitumor agents.

91. EFFECTS OF ANTICANCEROUS AGENTS ON THE SERUM PROTEIN AND CONJUGATED PROTEIN FRACTIONS OF PATIENTS WITH MALIGNANCY

SHIGEMI INUI and YASUYO SHIMIZU

(1st Department of Internal Medicine, Gifu Prefectural Medical School)

The effects of Tespamin, Myleran, Carzinophilin and Mitomycin C on the serum protein and conjugated protein fractions were studied in patients with malignant tumor by paper-electrophoretic method. The results were as follows:

- 1) Changes of the serum total protein and albumin fractions were not constant, but in cases which showed good clinical course during Tespamin treatment, the albumin values increased, compared with the pretreatment values.
- 2) α_1 - and α_2 -globulin had a tendency to return normal values during treatment with anticancerous agents.
- 3) β -globulin showed no constant changes.
- 4) γ -globulin decreased by Myleran and Carzinophilin administration.
- 5) Conjugated protein fractions approached to normal values.

92. THE METHOD DECIDING THE EFFECT MECHANISM OF ANTICANCER AGENT BY FLUCTUATION OF PHOSPHATASE VALUE IN ASCITES EXTRACTED FROM YOSHIDA SARCOMA RATS

MASAO OGIWARA, SHITAU YAMADA, SHIMAE TOSHIMA,
SUSUMU SHIMIZU, ISAO KATOO, TADASHI UMESONO,
FUJISHIGE NAKADA and NOBUO TANAKA

(Dept. of Yamada Internal Medicine, 3rd Hospital, Jikei Med. School)

The present method determining anticancer agent standard refers to its effect for animal life, morphological change in malignant tumour, number of cells and cell change *in-vitro*.

However, it is essential to make observation for malignant tumor and anticancer agent to react upon the living body with consideration of side action. Therefore we measured acid & alkali phosphatase value in ascites by utilizing rats transplanted Yoshida sarcoma.

When measuring P-ase, we adopted Seligman method using sodium β naphthol phosphoric acid as stroma.

Observation started from the fifth day after transplanting Yoshida sarcoma. Treatment was made every day for not only five cases of rats with the injection of Nitromin N oxide 10 mg/kg but also five cases of filtrate of some bacillus 30 mg/kg to abdominal cavity. 0.1 cc ascites was extracted from each group and measured its P-ase.

Score

On the first day, alkali P-ase was 22.7 γ /1ml on the average in the contrast case and acid P-ase 245 γ /1ml. Since the fourth day, both P-ase increased slightly and they amounted to one and a half of that on the first day.

In the group with Nitromin N oxide, remarkable increase of P-ase namely 15 times in alkali and 5 times in acid of that before injection was found from the second day through the third.

It became approximately equal to that before injection from the sixth through the seventh.

With treatment, number of cell increased remarkably.

In the group with filtrate of some bacillus, alkali P-ase started to increase from the fourth day and became respectively double on the sixth day and equal to that before injection on the eighth day. Acid P-ase increased to double that in the contrast case, but was often found to be less than that before injection. Yoshida sarcoma cell decreased with treatment.

As the result of the above investigation, it was noted that direct action of Nitromin N oxide upon Yoshida sarcoma caused remarkable cell destruction and increase of P-ase, and action of filtrate of some bacillus may be inflammatory reaction. Judging from the above statement, fluctuation of P-ase in ascites serves as an aid for finding the difference in function of the various anticancer agents.

93. STUDIES ON PROTECTION OF LEUKOPENIA DUE TO ANTI-TUMOUR AGENTS. (II) EFFECT OF SOME LEUKOCYTOSIS-PROMOTING SUBSTANCES ON LEUKOPENIA DUE TO RC-4

YOSHIHIKO INAZU, YUKIO SUGIYA, MASAO ARAKAWA
and SHIGEYUKI OSAMURA

(Takamine Laboratory, Sankyo co, LTD.; Dept. of Internal Med., Tokyo Medical Collage)

Previously, on the General Meeting of the Japanese Cancer Society last year, We reported effects on blood and bone marrow picture of leukocytosis-promoting agents

such as Fe-Chlorophyllin, Cytochrome C, L-Cysteine, β -Aminoethylisothiuronium (AET), β -Phthelimidethylisothiuronium (PT), DNA or Adenine in combination with *p*-Phenylendiphosphoric acid tetraethylenimide (RC-4). In this report, examinations were made on the effect of those agents in perfusion of bone marrow of the femur of rabbits, on oxygen consumption of bone marrow and upon the survival time of the Ehrlich ascites carcinoma-bearing mouse.

The results obtained are as follows:

1) In bone marrow perfusion of the femur of rabbits 4 days after intravenous injection of RC-4 at a dose of 30 mg/kg body weight, all test agents except Adenine, which caused no change, caused a very slight increase in leukocyte counts and in neutrophilic leukocyte counts, although no difference recognized between them.

2) Oxygen consumption increased in bone marrow cells of the femur of rabbits receiving Fe-Chlorophyllin, Cytochrome C or L-Cysteine following the same process as above, though no or slight change was observed in the cases with the other agents.

3) RC-4 and one of the test agents were intravenously injected to mice 3 days after transplantation of Ehrlich asites carcinoma at a dose of 10 mg/kg body weight / day for 10 days, followed by blood drawing and killing, and bone marrow picture was examined. Decrease in leukocyte counts and neutrophilic leukocyte counts observed in samples from animals receiving RC-4 alone was prevented by administration of the test agents and the effect was relatively great in cases with Fe-Chlorophyllin, Cytochrome C, AET, PT, DNA, and Adenine among the agents tested.

Present ratios of leukocyte and granulocyte cell ratio did not change after injection of RC-4 alone but increased by the combined use of the test agents.

The increase was relatively great in the cases with Fe-Chlorophyllin, Cytochrome C, AET, PT, and Adenine.

Summarized the data in parts I and II, Fe-chlorophyllin, Cytochrome C, AET, and Adenine are considered by us to be the most effective.

III. Cytology

94. A CRITIC ON NTS STAINING FOR CANCER CELLS

KAZUO MORISHITA and KOZO UTSUMI*

(Dept. of surgery and Cancer Institute*, Med. School, Okayama Univ.)

It would be a great help to distinguish the cancer cells from normal ones, if the former could be stained selectively by some method. In 1959 Donomae reported that by staining with NTS (Sumitomo Chemical Works) at pH 9.8 cancer cells gave a more marked fluorescence than normal cells. This has been reconfirmed by the authors demonstrating that the dye has a strong affinity to the nucleus of cancer cells. In this connection, for the purpose to reveal the staining mechanism of NTS, we observed the binding of the dye with nucleoprotein from cancer cell, Ehrlich ascites tumor cell, in the media of varied pH, the crude nucleoprotein from mouse liver, crystallized hemoglobin from horse blood, ovo-albumin and cow serum served as the controls.

The binding test, performed on the filter paper by dropping a mixture of the dye with each protein, revealed that the dye combined with nucleoprotein only at pH 10 and 11 but all the proteins show the affinity to the dye at lower pH, 2 and 4. The result suggested that the staining would be correlated with the I.E.P. of protein.

The paper electrophoresis on the serum dye mixture showed that the dye moved with proteins in media whose pH is higher than the I.E.P. of proteins demonstrating that the positively charged proteins only bind with the dye.

Quantitative estimation of basic proteins from both tumor cells and normal cells revealed that the cancer cells are superior to the normal cells in contents of the basic protein. This will explain the more marked fluorescence of cancer cell than normal cells by staining with NTS.

The vital staining test with NTS by which Donomae claimed cancer cells were stained specifically, proved that degenerated cells only can be stained but not the cancer cells.

95. STUDIES ON THE ASCITES HEPATOMA. XIX. FURTHER STUDIES ON THE CHROMOSOME

TOMIZO YOSHIDA, HIDEHIKO ISAKA, SHIGEYOSHI ODASHIMA,
MOTOI ISHIDATE and TAKASHI YAMADA
(The Med. Institute of Sasaki Foundation)

The ascites hepatoma, prepared by ascitic conversion of the aminoazodye-induced hepatoma of the rat, can provide a series of ascitic tumors for comparative studies on a variety of a common normal ancestral cell, the liver cell.

In eight different strains of the ascites hepatoma, i.e. AH 13, 66 F, 39, 414, 62 F, 99, 62 and 310, the chromosome was studied with special respects to the number of chromosomes as well as the chromosome morphology and was compared with each other.

1. Significant differences were noted in the modal value and the variation range of the chromosome of the eight strains as indicated in the table. Each strain presents one-modal variation, without suggesting the occurrence of admixed cell populations of different modal chromosomes.

Distribution of the Chromosome Number of Ascites Hepatomas
AH 13, 66F, 39, 414, 62F, 99, 62 and 310
(Examined on 4-day-old Tumors)

Strains	No. of nuclei examined	Mode	Range of variations
AH 13	100	38	36-39
AH 66F	100	38	35-41
AH 39	50	44	42-46
AH 414	100	46-47	44-51
AH 62F	100	48	45-51
AH 99	100	64	61-67
AH 62	100	67-68	64-70
AH 310	100	71-72	68-76

2. Distinct differences were also observed in the morphology of chromosomes of each tumor strain. Two or three prominent large metacentric chromosomes were noted in three strains, i.e. AH 13, 66 F and 99, out of the eight tumors examined with the frequency of more than 60%. A minute and a satellited chromosome were found in the nuclear plates of AH 310.

The results showed that each tumor, originating from a common normal ancestry, the liver cell, has its own chromosomal pattern.

This investigation was made possible through the support and sponsorship of the U.S. Department of Army, through its Far East Research Office.

附 議

吉田俊秀：(1) 発生のどのような時期で観察されましたか。

(2) 吉田内腫で J 形染色体をもつ細胞はそれをもたないものとの間に差異はないと申されましたがどのような質において差異はなかったかという点についておうかがいいたします。

井坂英彦：① 各系統の染色体検査は、どの系統もかなり長く累代移植をした後に実施した。

② われわれの吉田内腫は、3 年前には大きな丁字形染色体を 1 本持っていたが、現在はない。しかしその性質は、移植性、成長速度、一般形態、薬物感受性などに関するかぎり、少しも変化がみられない。染色体の形態とその腫瘍の性質との関連は、簡単にきめつけることはできない。

96. COMPARATIVE IDIOGRAM ANALYSIS OF MN-SARCOMA CELLS (T-STRAIN) MAINTAINED BY CONTINUOUS TISSUE CULTURE AND THOSE MAINTAINED BY SERIAL ANIMAL PASSAGES

MANABU TAKAHASHI, KATSUMI HAMADA, HIRONOBU AWAYA
and KAZUO INOUE

(Dept. of Pathology, Yamaguchi Med. School)

As previously reported by the authors, MN-sarcoma cells (Mori-Nishizuka) maintained by continuous tissue culture (TC-type) are less virulent in the native host than the cells maintained by serial animal passages (AP-type). This time, we made a comparative idiogram analysis of these two types of cells.

Materials and Methods:

MN-cells of AP-type were obtained from the ascites of NA 2 mice of the 176 th transplant generation. MN-cells of TC-type used for the analysis had been cultivated in medium YLA for 129 days (33 generations) or more. A cold hypotonic saline technique was employed for the expansion of the chromosomes. Colchicine was not used.

Results:

1) MN-cells of AP-type. In 71 metaphase plates examined, the chromosome numbers were distributed between 31 and 76. The cells with 38 chromosomes were most frequent, comprising about 17% of the population. Five cells (7%) belonged to the tetraploid group. The karyotypes varied. In general, the chromosomes were rough-surfaced and the chromatids well separated. There were two to five chromosomes which had the kinetochore at the subterminal region. They belonged to the longer or medium-sized chromosomes. All others were telocentric, metacentric ones being absent.

2) MN-cells of TC-type. In 86 metaphase plates, the range of chromosome number variation was between 34 and 95. The modal chromosome numbers of the diploid group were 41 and 42; each being represented by 11 cells (13% of the population).

Six cells (7%) were in the tetraploid region. The chromosome morphology showed a greater variation than in the AP-type. The chromosomes were smooth-surfaced and separation of the chromatids was incomplete. Besides the chromosomes with a subterminal kinetochore, there were two to four metacentric chromosomes. The distinct features in this type were a great variation of the thickness of the chromosomes and a frequent occurrence of segmented chromosomes. The frequency of V-shaped chromosomes was slightly higher than in the AP-type, and when present, it belonged to the longer group.

Conclusion:

- 1) MN-cells of AP-type are hypodiploid, and those of TC-type hyperdiploid.
- 2) There is a greater variation of chromosome morphology in the TC-type than in the AP-type.
- 3) These two types of cells show different characteristics in their karyotypes.

附 議

前田: neardiploid (二倍体) などの表現を用いなくて染色体の数をもって表現することを提案する。後日の文献として役立つと思う。

森 茂樹: 実験腫瘍部門において、近年核学の研究がさかんであって、本研究は核形などの他に核酸の研究と比較することも、腫瘍性——正常細胞の腫瘍化さらに腫瘍細胞の悪性度の変化——の課題すなわちそれが *mutation*, *secondary mutation* 等との解決に役立つものと認める。かかる研究には適切な腫瘍種を用いることが重要であるが、現在吉田肉腫や腹水肝癌等が用いられている。しかしなお特徴のある私どもの造った MN 系肉腫もひろく学界人に用いられることを希望する。

MN (森・西塚) 系肉腫は N_2 均一系マウスに京大にて合成したエストロゲン、アクリロニトリルを連続投与して造ったもので、淋巴組織より発生したもので、その性状は癌学会病理学会などに発表している。移植率はほぼ吉田肉腫と同事である。*in vivo* のみならず *in vitro* にも継続させ得、またただ今高橋君発表のごとく核学をその他の性状についても発表している。かかる根拠から一般に広く用いられることを願っている。

高橋: ここで、腫瘍細胞集団の染色体数に関する特徴を hypodiploid, hyperdiploid 等とよび、modal chromosome number でよばなかったのは、一般に染色体数の分散の大きい集団においては、mode を一つに確定しがたい場合が多く、例えば組織培養型 MN 細胞においても、mode は時期によって 41~43 の間を変動しているからであります。このように染色体数の分散の大きい細胞集団における染色体数の代表性として、mode を取らないで、mode から遠く離れた染色体数を持つ細胞をも考慮に入れた値として、median を取って表示してはどうかということを提案したいと思います。演者は時期をかえて抽出した標本においてそれぞれ mode が異なり、一つの mode を決められなかった場合においても、それらの median を調べると、よく一致した値が得られることを認めておるので、ここに追加しておきます。

勝田: 今日組織培養界で問題になっているのは、細胞を長期培養していると、大抵の場合最高頻度の細胞の染色体の数が変わってしまうことでむしろいかにして変らない細胞株を得るかということが重要なのである。

**97. STUDIES ON STRAIN HELA CELLS (CERVICAL CARCINOMA
OF HUMAN UTERUS) IN PROTEIN-FREE MEDIA. (II) COM-
PARISON OF CHROMOSOMES AMONG THE CELLS
CULTIVATED IN SERUM MEDIUM AND TWO
SUBSTRAINS IN PROTEIN-FREE MEDIUM**

HIDEO OKUMURA, TOSHIKO TAKAOKA*
and HAJIM KATSUTA*

(Dept. of Anatomy, School of Med. Toho Univ.)

(Tissue Culture Laboratory, Institute for Infectious Diseases, Univ. of Tokyo*)

In the first paper the continuous cultivation of the HeLa cells in a protein-free medium was enabled by the use of polyvinylpyrrolidone in place of serum proteins. And two different substrains, HeLa_{P1} and HeLa_{P2}, were established. In the present work the chromosomal composition was compared among the original HeLa and the two substrains.

As widely known, variation is easily apt to be induced among the strain HeLa cells. According to laboratory difference is not seldom observed in their chromosomal composition.

1) The most predominant cells among the HeLa cells, cultivated in the present laboratory continuously in the serum medium, show 76 elements of chromosomes.

2) Among the HeLa_{P1} and HeLa_{P2}, the 74-elemented cells were found to be predominant. And, similarly to the finding from the comparison between the L and L_{P1} cells, the distribution range in the number of chromosomes was also much narrower among the two substrains of the HeLa in the protein-free medium than that among the original HeLa in the serum medium.

3) The karyotype of most of the predominant cells among the HeLa in the serum medium contained 20 elements of V-shaped and 25 to 27 elements of J-shaped chromosomes.

4) The predominant cells with 74 elements among the HeLa_{P1} and HeLa_{P2} were almost similar in the chromosome complex to that with 76 elements among the HeLa cells.

This work was supported in part by the grants from the Ministry of Education, from the Ministry of Agriculture and Forestry, and from Asahi Shimbun Publishing Co.

**98. STUDIES ON STRAIN L CELLS (MOUSE FIBROBLASTS) IN
PROTEIN-FREE MEDIA. (VIII) EFFECTS OF SERA ON
THE APPEARANCE OF MULTINUCLEATED CELLS**

MAMORU KAWANA, KAZUE KAWAGUCHI, TOSHIKO TAKAOKA,
MAKOTO UMEDA and HAJIM KATSUTA

(Tissue Culture Laboratory, Institute for Infectious Diseases, Univ. of Tokyo)

Effects of the following nine kinds of sera were examined on the cell multiplication and on the appearance of multinucleated cells in tissue culture of the L_{P1} cells, a substrain of the L cells adapted to a protein-free medium containing polyvinylpyrrolidone on behalf of serum proteins: Bovine serum, horse serum, rat serum, two kinds of mouse sera (strains C_3H and CFW) and fourteen kinds of human sera. And considerable differences were found in the effects among the sera.

1: The appearance of multinucleated cells was accelerated by the addition into the medium in most of the sera examined, in the order of mouse serum (CFW), human sera (#2 and #3), mouse serum (C_3H), bovine serum, rat serum and horse serum. The frequency was 8.30% in the 4th day culture with CFW serum and 0.80% with horse serum, while it was 0.30% to 0.68% in the protein-free medium.

2: Large size of cells and nuclei were more abundant in the cultures with mouse sera, especially with CFW serum.

3: By the addition of horse serum cell aggregation was formed apparently in the culture and increasingly following the cultivation.

4: Individual difference was marked in the effects of human serum.

This work was supported in part by the grants from the Ministry of Education and from the Ministry of Agriculture and Forestry.

**99. STUDIES ON STRAIN L CELLS (MOUSE FIBROBLASTS) IN
PROTEIN-FREE MEDIA. (IX) COMPARISON OF CHROMO-
SOMES AMONG FOUR SUBSTRAINS CULTIVATED
CONTINUOUSLY IN PROTEIN-FREE MEDIA**

HIDEO OKUMURA, TOSHIKO TAKAOKA* and HAJIM KATSUTA*

(Dept. of Anatomy, School of Med, Toho Univ.)

(Tissue Culture Laboratory, Institute for Infectious Diseases, Univ. of Tokyo*)

The four substrains of the L cells cultivated continuously in different protein-free media, L_{P1} to L_{P4} , were compared of the number of chromosomes and the karyotype

to each other. In the 3rd paper little change was already reported in the main genetical characteristics of the most predominant cells between the original L and the L_{P1} cells. The present work revealed as follows:

1) The distribution range in the number of chromosomes was markedly narrower among the substrains in the protein-free media than among the L cells in the serum medium.

2) The chromosome number of the most predominant cells was 66 elements among the L_{P2} , L_{P3} and L_{P4} cells, being different from that of the L and L_{P1} cells, 68 elements.

3) The characteristics, however, of the karyotype of those 66 elements were not so different from those of the 68 elements among the latter, in showing 11 elements of V-shaped and 5 elements of J-shaped chromosomes except 2 elements of rod-shaped ones reduced in the formers. However, among the 66-elemented karyotypes in the L_{P4} , such types were also observed so as to contain 13 elements of V-shaped and 5 elements of J-shaped chromosomes or to show 8 elements of J-shaped ones.

4) The 66-elemented cells were also found among the L and L_{P1} cells, although not so much.

5) The karyotype of the 68-elemented cells among the L_{P2} , L_{P3} and L_{P4} cells resembled closely to that of the predominant cells among the L and L_{P1} cells.

This work was supported in part by the grants from the Ministry of Education, from the Ministry of Agriculture and Forestry, and from Asahi Shimbun Publishing Co.

附 議

吉田俊秀: (1) *in vivo* で L 株細胞の染色体数はどのように分布しておりますか。御発表では 68 個のみと報告されたようですが。

奥付: 1) L 株細胞で predominant の 68 本の染色体の核型は一種類だけではありません。

2) L 株細胞は組織培養株でありますからその状態はわかりません。

IV. Histology

100. STUDIES OF INCLUSION BODIES IN HUMAN CANCER CELLS

KIN'YA OKANO, KIYOKAZU NAGAI, HIROTSUGU UDA,
YOOICHI MORI, TAKEO SAKI*, HARUO TANIGUCHI,
GAIKOO UEDA, TSUNEHICO FUJIMOTO and FUMIHIKO TARUTANI**

(2nd Dept. of Path. Med. School Osaka Univ.*)

(2nd Dept. of Path. Wakayama Med. College**)

Four cases of malignant tumors, which had inclusion-like bodies in tumor cells, were studied.

No. 1. An autopsied case of male aged 69 yrs. Pathological diagnosis was hepatic cancer (carcinoma hepatocellulare) with liver cirrhosis. Histologically, a marked cellular atypia with large nuclei and nucleoli was seen. Some of the inclusion bodies were strongly positive in PAS reaction, but showed no change after salivary digestion. By Mallory-Azan staining, these were violet-colored and metachromasia reaction with gentiana violet was positive. In Unna-Pappenheim staining inclusion bodies were pyronophil.

No. 2. A female, 34 yrs. of age, died from mammary cancer (adenocarcinoma) with widespread generalized bone metastasis, was autopsied. Prominent rounded inclusion bodies were observed in the cytoplasm, which were strongly positive in PAS reaction and did not show negative change after salivary digestion. Also, these were pyronophil, positive in metachromasia reaction and light blue colored in Mallory-Azan stain.

No. 3. Autopsy was performed on a female, 48 yrs. old, died from gastric cancer (carcinoma simplex). Pyronophil and PAS positive rounded bodies were observed in the cytoplasm of cancer cells. After Mallory-Azan staining, these were violet in color.

No. 4. A biopsied case of skin carcinoma of female, aged 58 yrs. Histological diagnosis was undifferentiated epidermoid carcinoma. A large number of cytoplasmic inclusion bodies, scattered in cytoplasm of tumor cells with halo formation, were observed. All of them were negative in PAS reaction, but pyronophil in Unna-Pappenheim staining. By Mallory-Azan staining, these showed violet red in color tone. Negative reaction in metachromasia staining was seen. Tissue culture was performed in this tumor, i.e. 4 times of the subculture were succeeded and its survival period

was about 50 days. Moreover, inclusion bodies were proved in cell colonies. Electron microscopically a large homogenous inclusion-like body of high density was seen in the cytoplasm and large vesicle, which contained some granules about 15 m μ in diameter.

Conclusively we are of the opinion that these inclusion bodies may be an expression of dysfunctioning stage in the so-called functioning tumors, which may or may not be caused by viruses.

101. NEW INTRACYTOPLASMIC AND INTRANUCLEAR CRYSTALLIZATIONS IN VARIOUS TUMOR CELLS

ZENSUKE OTA and HIROSHI MATSUMORI

(2nd Dept. of Internal Medicine, Medical School, Okayama Univ.)

By phase contrast microscopy, hitherto unknown intracellular crystallization were discovered to appear within the cytoplasm and nuclei of various types of human normal and tumor cells on slow cellular degeneration, in supravital preparations kept standing at room temperature up to ten days.

Bone Marrow

Some of normal neutrophils showed moderately refractile, hair-like crystalloids and rod-shaped, moderately refractile crystals in the cytoplasm.

Megakaryocytes displayed rod-shaped, highly refractile intracytoplasmic crystals in a case with monocytic leukemia.

In acute myelogenous leukemia, some of leukemic cells revealed two types of intracytoplasmic crystals. One was large, fusiform and moderately refractile, and the other short, rod-shaped and highly refractile. These crystals were also seen extracellularly. Similar types of intracytoplasmic crystals were observed in chronic myelogenous and monocytic leukemias. Rod-shaped, moderately refractile intranuclear crystals were seen in chronic myelogenous and monocytic leukemias.

In acute lymphocytic leukemia, rod-shaped, highly refractile crystals arose on the cytoplasmic margin of the leukemic cells and in chronic lymphocytic leukemia similar crystals in the mass of the degenerated cells.

Pleural Effusion: Neutrophils in pleural effusion exhibited rod-shaped, slightly refractile intranuclear crystals in a patient with breast cancer.

Ascites: A few of ascitic phagocytes showed rod-shaped, highly refractile intracytoplasmic crystals. In celothelioma, the tumor cells exhibited rod-shaped, moderately refractile intracytoplasmic crystals of varying size and large, rod-shaped,

moderately refractile intranuclear crystals. In carcinomatous peritonitis due to gastric cancer, the tumor cells displayed similar intracytoplasmic and intranuclear crystals of smaller size.

Stomach Tissue: Some of cells obtained from stomach mucosa and stomach cancer tissue showed rod-shaped, highly refractile intracytoplasmic crystals.

In conclusion, intracellular crystallizations seemed to show some different characteristics in leukemic cells from those in normal blood cells and were much more frequently encountered in tumor cells.

102. INVASIVE ABILITY OF DIPLOID AND TETRAPLOID TUMOR CELLS IN RATS AND MICE

TOSHIHIDE H. YOSHIDA (Dept. of Cytogenetics, National Institute of Genetics)

In order to investigate the nature of invasive ability of tetraploid tumor cells, the following experiments were carried out with Yoshida and Ehrlich ascites tumors. Yoshida rat sarcoma used in the present study was characterized by tumor stem cells having near-diploid chromosome numbers (± 40 chromosomes), but 7.3% of its cells were found to have near-tetraploid chromosome sets. Tissue pulps of kidney, spleen and thymus of the tumor bearing animals were transferred into the peritoneal cavities of new hosts. As a result the tumor cells which invaded the organs developed into ascites tumors. Two ascites tumors developed by spleen transplantations showed 33.3 and 12.0% tetraploid cells. One ascites tumor by transplantation of kidney was characterized by tetraploid cells at 19.0%, while two ascites tumors by thymus transfer resulted to have tetraploid cells at only 2.5 and 5.3%.

A near-diploid Ehrlich ascites tumor which contained tetraploid cells at 13.0% was also used to investigate the invasive ability of tetraploid cells. Lung tissue of a tumor bearing animal was transferred into the peritoneal cavities of mice. Two ascites tumors developed by the lung transfer were characterized by having tetraploid cells at 71.6 and 30.9%, respectively, while in the control animals they amounted only to 11.0%.

By intravenous injection of the near-diploid Ehrlich tumor two mice developed tumors. One of them was developed the tumor under the skin in the upper part of the ventral side and the other developed two tumor masses on the dorsal side and in the kidney. These tumor pulps were injected into the peritoneal cavities by usual transfer technique. Four ascites tumors which developed by transplantation of the ventral tumor showed 85.0, 47.3, 57.0 and 57.8% tetraploid cells, respectively. One

ascites tumor developed by inoculation of the dorsal tumor was characterized by tetraploid cells at 40.4%, while the kidney tumor transfer resulted in two ascites tumors containing tetraploid cells at 22.6 and 11.6%, respectively. Based on the above investigations it seems that tetraploid tumor cells invaded the organs easier than the diploid ones.

103. ON THE AGE FACTOR ON THE HISTOLOGICAL VARIATION IN THE CANCER TISSUES WITH SPECIAL REFERENCE TO THE RELATIONSHIP BETWEEN PICTURES OF CANCER CELLS AND OF INTERSTITIA

TSUNEKO SATO and HISASHI TAUCHI

(2nd Dept. of Path, School of Med., Nagoya Univ.)

One hundred and twenty seven autopsy cases of primary lung cancer (111 male, 16 female) were histologically examined with special reference to the morphological character of its interstitia.

The pattern of the appearance of various cancer pictures was considered to be influenced by the environmental condition of cancer growth and some interesting findings were obtained in the pattern of appearance of some cancer histology with advancement of age.

It was considered that the most characteristic picture in old age was highly differentiated squamous cell carcinoma forming larger cancer alveoli with fine fibrillar or loosely fibrous interstitium, and in younger cases high columnar typical adenocarcinoma with various types of interstitium. In case of younger age, there was generally seen marked proliferative picture of the stroma in which cancer cells formed smaller alveoli.

In general, the picture of typical adenocarcinoma were more frequently seen in female cases than in male cases. In this case, the histological variation was somewhat poorly recognized. In case of lung cancer, picture of typical adenocarcinoma may be discussed histogenetically on another point of view from other cancer histology.

The histological pictures were considered to show that the cancer cells grew slowly with a picture of more complete differentiation of the cells in old age and rapidly with a picture of less complete differentiation in younger age. It was found that some of cancer cells especially in case of picture of non-typical adenocarcinoma became smaller in size in older age.

It may be considered that variation of the character of cancer cells according to

age was due to different changing process of the interstitium which influenced by age factor in the course of cancer growth.

附 議

今井 環：癌細胞のいわゆる簇出發育部での間質結合組織増殖は癌細胞の退行像と関係が深いことが少ないと思うが、御所見を御教示いただきたい。なお、退行過程というのは、癌細胞の萎縮状のものから、形態不同、壊死など、かなり広くとった形態学的所見のつもりである。

佐藤：私どもが癌細胞と間質との形態学的関連性を検討します際には、その場において癌細胞が変性壊死の過程に陥入らず、一応増殖していたと考えられる部についてのみ、検討しその概略を集計して考察を加えました。

田内：いろいろと御教示いただきたいことがたくさんありますが、私どもの研究の経過をゆっくり検討していただきたく存じます。

104. HISTOLOGICAL STUDIES ON MUCOSAL CARCINOMA OF THE STOMACH. FUNDAMENTAL STRUCTURES OF THE CANCER TISSUES AND THEIR RELATIONSHIP TO THE HISTOLOGICAL PICTURES OF THE NON-CANCERIZED GASTRIC MUCOSA

TAKEO NAGAYO and TAKASADA KOMAGOE
(2nd. Dept. of Path. Med. School, Nagoya City Univ.)

As is well known, histological picture of advanced gastric carcinoma are varied even in a case. This seems partly to be due to the influence of local or general factors, which occurred secondarily in the patient by growing of the carcinoma. Therefore it is considered that gastric mucosal carcinomas, which are less influenced by these factors, are relatively simple in histological pictures and relationship between histological pictures of the mucosal carcinoma and of the remaining non-cancerous pyloric mucosa are not so complex as in cases of advanced carcinoma.

From these standpoints of view, 42 cases of surgically removed gastric mucosal carcinoma (28 cases of ulcer-cancer; I group, 5 cases of polyp-cancer; II group, and 9 cases of superficially-spreading erosive carcinoma without ulcer; III group) were examined histologically and following results were obtained.

1. Histological pictures of mucosal carcinoma were classified into following three types. Namely, A) Adenocarcinoma (subdivided into papillary-, tubular- and acinar Adenocarcinoma) B) Fascicular or reticular carcinoma (composed of solid fascicles of cancer cells and showed little or no glandular structure) and C) Dissociated cell carcinoma (composed of signet-ring cancer cells)

2. In many cases (43%) transitional structure was recognised between A type and

B and B type and C.

3. Regardless of gross appearances of mucosal carcinoma, almost all cases of A type showed severe or moderate grade of intestinal metaplasia in the remaining pyloric mucosa. On the contrary, metaplasia were noticed scarcely in cases of C type. Cases of A+B type had higher grade of metaplasia than cases of B+C type. Namely, intimate relationship was confirmed between histological pictures of mucosal carcinoma and grade of intestinal metaplasia in the pyloric mucosa.

4. All cases of polyp-cancer showed a picture of A type especially papillary or papillo-tubular adenocarcinoma. All cases of superficially-spreading erosive carcinoma showed A or A+B types except one case of B+C type. Ulcer-cancer, however, showed no these histological characteristics between cancer tissues and non-cancerous pyloric mucosa.

附 議

友田：御発表の要旨と多少かけはなれるかも知れませんが、胃粘膜癌症例の転移の様相についておうかがいいたします。

早期胃癌を手術の対象としながら、外科医が癌であることを明確に認識し得なかった場合には、慢性胃炎等と考えて胃切除を施すと、転移廓清が不十分となり、治療成績が、早期胃癌でありながら却て悪い結果にもなり得ます。従て胃粘膜癌の組織学的研究に当っては、同時に転移についての研究が必要だからであります。

長与：転移についてはふれなかったが、42 例中の 2 例（いずれも潰瘍癌組織像は B-C, C 型）の幽門輪直下リンパ節に鏡鏡的な微少転移巣をみている。

太田邦夫：①潰瘍癌の場合 A, B, C 型の増殖と附近の粘膜の化生とは一応関係がないと述べられたと思いますが私どもも同様の所見を得ております。この所見といわゆる潰瘍癌および胃炎の組織発生との関係をどのように御解釈になりますか？

②いわゆる潰瘍癌には二種あって、一つは事実潰瘍辺縁に生ずるものであり、他は辺縁から多少はなれて発生するものであるという御考えでしようか？

長与：潰瘍癌には発生的に二種類があると思う。一つは潰瘍辺縁の再生粘膜上皮から発癌したと思われるもので、他は潰瘍辺縁粘膜から数 mm はなれた粘膜に癌の見られるものである。後者の場合はビラン型をとるものが多く、再生上皮から出たものか否かははっきりしない。

今井 環：腸上皮化生の程度の判定は、胃のどの範囲の所見によったか。

105. A CLINICO-PATHOLOGICAL STUDY OF EARLY ULCUS-CARCINOMA

MAMORU SOMEYA and KUNIO TAKAGI

(Department of Surgery, Cancer Institute Hospital)

Among 1913 resected cases of stomach-cancer, from 1946 through 1959, 153 were recognized as ulcer-carcinoma by histological examination. There were 60 cases which apparently represented early developmental stage of ulcer-carcinoma.

In 31 of them the mucosal layer was the exclusive site of location (1 group) and in 29 the submucous layer also involved only to a limited extent (2 group). Their ratio to total gastric ulcer cases were 3.1% and 6.3% respectively. It should be emphasized that a sharp increase in such cases has been noticed in the recent six years.

Age distribution showed remarkable predominance in the 4th decade. As to the sex-ratio, early ul-ca. associated with deep penetrated ulcers were much more frequent in male ($\sigma : \phi = 4.5 : 1$), while in cases with ulcers not involving the muscularis propria, both sexes were equally involved. The early ul-ca. tends to be localized in the intermediary zone along the lesser curvature. Further preponderance of the corpus-region of such lesions as compared with cancers and ulcers in general was noticed. In gross appearance the ulcers were round (85%) or linear (15%), and deep, shallow or cicatrizing ulcers were respectively 26.5%, 38.3% and 16.6%. Almost all of them showed mucosal cancer with regenerating epithellium around the ulcer, though Hauser said that cancer was seen at one side of ulcer in early stage. Very extensive mucosal spread associated with shallow ulcers or ulcer-scars was observed frequently. Many cases of intramucosal spread showed concentric spreading. As Takagi's report on the mucosal cancer at the 18th Cancer Congress, the tubular type of mucosal cancer was 56.7% in total cases but in ul-2 and ul-3 ca., about 47% was signet-ring cell type. Almost all cases of early ulcer-carcinoma showed atrophic metaplastic gastritis, except 4 cases of hyperplastic gastritis, one of which showed remarkable nodular hyperplasia at the pyloric edge of mucosal cancer. The submucosal infiltration was classified into 4 types; there were scirrhus, sprouting, lymphogenous and local types. The ul-2 and ul-3 carcinomas showed scirrhus or sprouting infiltration, while the ul-4 carcinoma took every growth type.

In reference to histological evidences of ulcer origin in early ul-ca. are regenerating epithelium, scar-tissue and intramucosal spread.

The frequency of lymph node involvement were 19.4% in group 1 and 24.1% in group 2.

The majority of the cases showed the symptom of ulcer-like type. Tumor was palpable only in 13%: blood reaction in stool was negative in half cases: achylia in 11.7%.

In 41.7% of cases malignancy was suspected by X-ray examination but differentiation between benign ulcer and ul-4-b-ca. in early stage was quite difficult.

The average duration of the disease was 1 year and 9 months, and rather long than gastric cancer.

The five-year survival rate of 19 cases from 1946 through 1955 was 89.5%, while the rate of advanced ul-ca. was 44.4%.

附 議

田崎勇三：染谷君の発表に敬意を表するものである。1913 例の胃癌切除例中の 153 例すなわち 8.0% が潰瘍癌であったがその中 60 例が比較的早期のものであった。しかも最近 6 年間にかかる早期の潰瘍癌が急激に増加を示したことを強調されたが、これは X 線診断技術の向上もあづかって力があったし、大衆の理解も進んだ結果と思われる。

腫瘍がふれるものが僅かに 13% で、大便潜血反応が半数において陰性を示したこと、無酸症は僅かに 11% であったこと、また 41.7% が X 線検査によって癌を疑われたことなどは特記すべきことである。さらに最近 9 年間の 19 例中 5 年治癒が 17 例すなわち 89.5% であることは、独り日本ばかりでなく、世界にも誇示すべき優秀な治療成績で賞讃に値するものである。

106. STUDIES ON GASTRIC CARCINOMA WITH REFERENCE TO THE RELATIONSHIP BETWEEN THE HISTOPATHOLOGICAL NATURE OF THE CARCINOMA AND THE ALTERATION IN THE CELLULAR CONSTITUENT OF THE REGIONAL LYMPH NODES

HUMIO KITADE, TETSUGO MORIOKA, TAMOTSU ISOHASHI,
KATSUJI FUKUDA, KATSUTOSHI HAYASHI and SAKAE ASADA
(Dept. of Surgery, Osaka Med. College)

All of the visible lymph nodes in the region of the stomach obtained at surgery were examined in 9 cases of gastric carcinoma in an effort to make clear the relationship between the histopathological nature of the carcinoma and the alteration in the cellular constituent of the regional lymph nodes.

Imprint specimens of lymph nodes stained by May-Giemsa and Papanicolaou methods were made and 2,000 cells were counted at random from the cortical portion. The percentages of various component cells were calculated.

It was clarified in this study that the percentage of lymphoblasts and reticulum cells in the regional lymph nodes from cancer cases were increased remarkably as compared to that of control specimens taken from gastric ulcer. This trend was more remarkable in the cases which showed disseminated non-continuous infiltration of cancer cells in the stomach wall than in those which showed continuous blanch-like infiltration. Furthermore, this trend was more remarkable in the lymph nodes in which metastases of cancer cells were proven, by tissue sections, than in those without metastasis.

When the lymph node metastases were divided into a group with metastatic cells restricted within the marginal sinuses and a group with the metastatic cells invading as far as the medullary portion of the node, the former contained more lymphoblasts while the latter was more abundant in reticulum cells.

The lymph nodes free from metastasis were also investigated. Here, lymphoblasts were more numerous than reticulum cells in the proliferated form with increase of lymphatic tissue. The opposite was true in the atrophied form, in which lymphatic tissue was atrophied.

107. A HISTOCHEMICAL STUDY OF THE GASTRIC CANCER: ACTIVITY OF SUCCINOXYDASE, DPN-DIAPHORASE, TPN- DIAPHORASE, α -GLYCEROPHOSPHATE DEHYDROGENASE AND PHOSPHORYLASE IN CANCER CELLS

TAKURO OGATA and MOTOO KITAMURA

(Dept. of Surgery, Med. School, Okayama Univ.)

A histochemical study of five metabolically important enzymes has been studied in the gastrectomy specimens of 42 patients. For the demonstration of succinoxidase (SD), diphosphopyridine nucleotide diaphorase (DPN-D), triphosphopyridine nucleotide diaphorase (TPN-D) and α -glycerophosphate dehydrogenase (α -GD), the modified method of Wattenberg was used and for phosphorylase (P-ase) the method of Takeuchi.

The average activity of all 5 enzymes in normal mucosa and cancer cells showed in the bellow table, but in a single specimen some variations of activity were observed. The cells at the periphery of tumor cell nest, at the invading margin and in isolated cell groups showed higher activity of DPN-D and TPN-D, while those at the central part showed lower activity of all 5 enzymes. From the present histochemical study, it was observed that in gastric cancer cells DPN- and TPN-diaphorase systems showed higher activity, while glycolysis, respiration and fatty acid metabolism lower.

	*SD	*DPN-D	*TPN-D	* α -GD	P-ase	Cases
Normal surface epithelium	1	6-7	6	1	+	5
Normal mucous neck cells	3	5	5	2	—	5
Normal parietal cells	6-7	1	1	6	—	5
Normal zymogenic cells	2	1	1	2	—	5
Well differentiated adenocarcinoma	4-5	5-6	5	3	+	12
Poorly differentiated adenocarcinoma	4	6	5-6	3	+	14
Carcinoma simplex	3	6-7	5-6	1-2	—	11
Scirrhus	2	6-7	6	1	—	5

* The number shows the average degree of neotetrazolium deposition, 7 maximum formazan deposition, 6 deep purple, 5 purple, 4 light purple, 3 red, 2 pink, 1 light pink.

And this tendency was more markedly observed in the poorly differentiated cells than in the well differentiated cells.

108. PATHOLOGIC-ANATOMICAL STUDIES ON CARCINOMA OF THE PANCREAS, WITH SPECIAL REFERENCE TO DIABETES MELLITUS AS A COMPLICATION

TAKUZO ISHIDATE (Dept. of Path., School of Med., Tohoku Univ.)

Primary carcinoma of the pancreas was found in 50 among 8877 autopsies performed at the Department of Pathology, Toohoku University School of Medicine during the last 45 years.

Of the total cases 36 were male and 14 female. The peak of the age incidence was in the sixth decade in male and the seventh in female.

The tumors were classified into 3 major types histogenetically. Among 39 cases examined, there were 26 of ductal carcinoma including 2 squamous carcinoma and one mucinous adenocarcinoma; 11 of acinar carcinoma, in one of which a part of isletcell-like carcinoma was intermingling; and 2 of indeterminate origin, where the tumors were composed of small carcinoma cells.

The initiation of the tumor was located in the head of the pancreas in 33 cases, distributed in the body and tail in 16 cases, and the tumor extended diffusely in the whole organ in one case.

No correlation was found between the location and the histological types.

Frequency of tumor spread and metastasis in the liver and duodenum was more marked in the cases of the headcarcinoma than in those of the body and tail, while metastasis into other organs rather high in the latter. Incidence of metastasis seemed to be correlated with the histological types, i.e., it was higher in cases of the acinar than in the ductal type.

It is generally accepted that hyperglycemia or glycosuria is sometimes accompanied with pancreatic carcinoma. In our materials, 8 patients had glycosuria prior to or after the onset of the disease. In all 6 of these 8 cases which could be examined histologically, carcinoma of the ductal type was observed. In these cases, the tissues of the islets were destructed markedly by tumor invasion with fibrosis. In cases of the ductal type without glycosuria there was observed the remnance of the islets, not affected by tumor invasion, but inbedded in a marked fibrosis showing their hypertrophy and hyperplasia. These features could be detected in cases of the acinal type, where none of glycosuria was observed. Among the cases examined,

there could not be found any case where the true diabetes mellitus had existed prior to the occurrence of pancreatic carcinoma. (文部省科学研究費による)

109. STUDY ON THE LYMPHATIC FOLLICLE OF THE UROLOGICAL TUMORS

TOKUJI KATO, JOJI CHIDOI and TADASHI AOKI

(Urological Clinic, School Med., Hiroshima Univ.)

Histological studies on the appearance of lymphatic follicle have been carried out on 322 cases with urological tumors. These consisted of 63 cases with renal tumor (39 cases of Grawitz's tumor, 12 cases of papilloma of pelvis, 7 cases of carcinoma pelvis, 5 cases of renal sarcoma), 86 cases with vesical tumor (the majority of them were cancer in nature), 118 cases with prostatic tumor (85 cases of prostatic hypertrophy, 32 cases of prostatic cancer), 31 cases with urethral cancer and 24 cases with testicular tumor.

The marked increase in the appearance of lymphatic follicle has been observed in the above series comparing the normal healthy subjects. The most significant increase in the appearance of lymphatic follicle was found in the patients with urethral tumor followed in the order of prostate, testicle, kidney and bladder.

110. HISTOPATHOLOGICAL RELATIONSHIP BETWEEN CANCEROUS AND MASTOPATHIC BREASTS IN JAPANESE WOMEN

MASAO FUJIMORI and MASARU IZUO (Mitsui Kosei Hospital)

Histopathological features of breasts have been investigated in detail on 598 mammary cancerous and 107 mastopathic patients past during the 6 years.

In mastopathic breasts blunt duct adenosis, cyst, apocrine epithelium and duct papillomatosis are found very often, on the other hand in cancerous breasts atrophy, fibrosis, blunt duct adenosis and duct papillomatosis are found very often (Table 1).

Duct papillomatosis appear in 30% to 40% of both diseases and are most often found in breasts of both diseases of the patients at 30 and 40 decades.

Summarized reports on the histopathological relationship between cancerous and mastopathic breasts from 40 surgical and 8 pathological departments of different universities in Japan are as follows (Table 2).

Table 1 Frequency of histological changes in cancerous or mastopathic breasts

Histological Changes	Mammary Cancer 598 Cases	Mastopathy 107 Cases
Cyst	16.9	58.9
Duct papillomatosis	31.8	37.4
Blunt duct adenosis	35.4	86.9
Sclerosing adenosis	16.6	33.6
Apocrine epithelium	26.3	56.1
Lobular hyperplasia	6.0	4.7
Duct epithelial hyperplasia	14.2	21.5
Atrophy	59.5	3.7
Fibrosis	53.3	28.9
Duct stasis	32.6	22.4
Fibroadenoma	10.5	5.6

Table 2 Histopathological relationship between mammary cancer and mastopathy

	Reporters	No. of Cases	%
Concomitant early cancerous lesions in the breasts of mastopathy	Fujimori	11/106	10.4
	Surgical Deptt.	104/1813	5.7
	Pathological Deptt.	69/680	10.1
Concomitant mastopathic lesions in the breasts of carcinoma	Fujimori	5/28	17.9
	Surgical Deptt.	222/713	31.1
	Pathological Deptt.	49/502	9.8
Carcinomatous alterations of mastopathy	Fujimori	5/106	4.7
	Surgical Deptt.	114/1875	6.1
	Pathological Deptt.	29/714	4.1

The frequency of concomitant early cancerous lesions in breasts of mastopathy is 5.7% to 10.4%, the frequency of concomitant mastopathic lesions in breasts of carcinoma is 9.8% to 31.1% and the frequency of carcinomatous alterations of mastopathy is 4.1% to 6.1%.

(Supported by the Scientific Research Grant of the Education Ministry)

附 議

今井 環：癌塊の大きさにより，例えば乳腺症性病変との合併頻度など，多少ちがってくるかと思うが，御検索の乳癌塊の大きさを，具体的に御教示ねがいたい。

藤森正雄：初期癌というのは臨床的に 0.5~1.0 cm 径の小腫瘍を指していったのであって，病理組織学的の厳密な意味からではない。

111. 2 CASES OF SPONTANEOUS MULTIPLE MAMMARY TUMORS WITH THEIR METASTASIS IN CATS

FUMITOMO WATANABE, IKUMI KUWATSUKA, SHIN OGATA
and MIDORI HASHIGUCHI

(Animal and Medical Institute, School of Med., Nagasaki Univ.)

人間ときわめて密接に生活する猫の乳腺腫瘍については最近佐伯氏等の報告がある。われわれは猫の乳腺部に発生した、癌腫、肉腫およびその転移を多発した自然発生腫瘍の2例を剖検したので報告する。

第1例：16才，♀，毛色は三毛，栄養良好，最近2回の分娩後授乳を好まず仔は全部死んだ。約1年前に右側第3乳腺部に腫瘍の発生を認めた。解剖時体重1680g，右第3乳腺；乳腺部に2.0×3.0×3.1cm，腺癌，管腔の發育は明瞭で均質な蛋白物質を容れる壊死巣散在。左第3乳腺；2.0×1.6×0.9cmの結節，潰瘍状。これに接してエンドウ大結節1個，細網肉腫ならびに腺癌。本来の乳腺上皮は増殖性。右第1，第2，第4，左第1，第4乳腺はいずれも萎縮性。左第2乳腺の上皮の一部は異型増殖。左鼠蹊部淋巴腺；転移結節2.0×2.5×0.5cm，細網肉腫ならびに腺癌の転移。細網肉腫像の中に腺癌の明瞭な構造があってその周囲には単純癌に近い像が見られたが多くの部分において定型的な細網肉腫像を示す。異種腫瘍の重複転移を思わせる。右鼠蹊部淋巴腺；2.0×1.1×1.0cm，細網肉腫転移。後腹壁淋巴腺；5.0×1.1×2.0cm，細網肉腫。頭部皮膚結節；1.8×1.4×1.0cm，蜂窩状黒色腫。

第2例：15才，♀，黒白，栄養良好，10回以上出産したが授乳を好まず，仔は發育しなかった。約半年前に右側第4乳腺に拇指頭大の結節が現れ，これを摘出，その後2ヵ月して死亡，解剖時体重1600g。右第1乳腺；2.9×2.6×2.1cm，腺癌ならびに多核巨細胞肉腫，明瞭な腺癌組織の部分と多核巨細胞を混える大円形核の肉腫状の部分からなる。右第3乳腺；2.6×1.6×1.7cm，腺癌，管腔形成明瞭，壊死巣散在。右第4乳腺；2.6×3.2×3.0cm，比較的長い円柱状の細胞が管腔を作るが充実性増殖部では円形の細胞よりなる。左第1乳腺；2.2×3.1×1.7cm，乳嘴腺癌，大きな明るい核の細胞（染色体数52以上）が明瞭な境界なしに連続的に増殖し，はなはだ不完全な腺管腔を作りその中に乳嘴状に突出。網眼状をなす間質結合組織が増生。左第1乳腺部小結節；腺癌，小型の上皮性の腫瘍細胞が大小の明瞭な管腔を作り壊死巣多し。これは第1乳腺内に発生した別の種類の腺癌とみなされる。左第4乳腺；米粒大，腺癌，大小の腺管腔を作って増殖，まれに充実性増殖部あり。右第2乳腺；萎縮性，異型増殖部あり，左第2，第3乳腺組織；萎縮性。転移；両肺の全葉にわたって粟粒大結節多数。1～

20 個の肺胞に腫瘍細胞を満たし腺状管腔にはコロイド様物質を入れる。膵、肝、脾、腎周囲ならびに後腹壁淋巴腺に腺癌転移あり。

考察：① 最近出産後授乳をしなかった栄養良好な2匹の老猫（16才，15才）に発生した多発性乳腺腫瘍とその転移について記述した。② 乳腺腫瘍の幼若猫への皮下移植は両例とも成功しなかったがウィルス性の起原は否定できない。③ 腫瘍のない乳腺には両例とも明瞭な前癌性増殖は認められなかった。④ 各乳腺癌の間には多少の構造上の差異はあったが内臓転移はどの乳腺癌から由来したものかは決定できない。⑤ 同一乳腺に癌腫と肉腫が発生し、淋巴腺に癌腫と肉腫の重複転移が起ることを示した。

112. STUDIES ON THE ATYPICAL PROLIFERATION OF CERVICAL EPITHELIUM AND CERVICAL CARCINOMA

NOBUJIRO TAKIZAWA and BIN TAKEDA

(Dept. of Pathology & Dept. of Gynecology, Medical School, Chiba University)

From 1955 to 1959, radical hysterectomy was performed in 335 cases in Gyn-Obst dept, Chiba Univ. Hospital. The authors found, among those operated specimens, 35 cases of atypical cervical epithelium. They consisted of atypical proliferation of squamous epithelium (22) and of cylindrical epithelium (13). The former was divided into glandular extension type (16) and papillary type (6).

Glandular extension of atypical proliferation of cervical squamous epithelium was compared with that of cervical squamous carcinoma. The parenchymal cells of the latter were more polymorphic, had more basophilic-protoplasm and more hyperchromatic nucleus membrane than the former. Besides, mesenchymal spindle-shaped cells close to the basement membrane in the latter were more likely to be degenerative, having swollen nucleus, while those of the former were not degenerative but proliferative.

Silvering stain revealed that argentaffin fibrils of basement membrane of the latter looked more irregular and lacked fine fibrils, compared with the former.

Such findings can serve to differentiate so-called glandular involvement.

113. HISTOCHEMICAL STUDY ON CARCINOMA IN SITU OF THE UTERINE CERVIX

TAKANOBU KAWANISHI

(Dept. of Obst. and Gyne., Med. School, Okayama Univ.)

Histochemical observations on RNA and DNA were done in the benign, precancerous tissues of the human uterine cervixes.

1) RNA: The methylgreen-pyronin Y solution was used for the determination of RNA, by means of the Taft's modification. The pyroninophilic nucleoli were assayed biometrically in 30 normal, 5 atypical, 5 precancerous and 30 cancerous epithelia. A similar nucleolar activity was found between both groups of carcinoma *in situ* and atypical epithelium. The mean diameter of single nucleoli in the both groups was significantly smaller than that in invasive cancerous group and was resembled to that in normal group. The number of nuclei with multiple nucleoli in both groups was similar to the invasive cancerous group.

2) DNA: DNA distribution of the squamous epithelia stained by Feulgen reaction was determined using 5600Å under the microspectrophotometer. The epithelia were crossed by the light spot at the diameter of 20μ, from basal to superficial layers. The DNA absorption curves at the structure of the cell layers were obtained in 20 cases of normal epithelia, 3 carcinoma *in situ* and 5 carcinoma *in situ* in the vicinity of invasive carcinoma. The absorption intensity from parabasal to superficial layers in the both groups of carcinoma *in situ* and carcinoma *in situ* in the vicinity of invasive carcinoma was higher than in the normal group. The quadratic absorption curves were obtained in the both groups of normal case and carcinoma *in situ*. On the contrary, an irregular curve was shown in carcinoma *in situ* in the vicinity of invasive carcinoma. It is considered that the DNA contents in carcinoma *in situ* are gradually decreased from basal to superficial layers, but in carcinoma *in situ* in the vicinity of invasive carcinoma, a slight difference of DNA contents are demonstrated between basal and superficial layers. (文部省科学研究費による)

114. HISTOLOGICAL CLASSIFICATION FOR STAGING OF CERVICAL CANCER

SHOJI IWAI, TAKESHI SAITO, KYUYO SHIOZAWA
and TAZUO TSUDA

(Dept. of Obst. and Gyne., Faculty of Med., Shinshu Univ.)

子宮頸癌の進行度分類としては臨床的分類が今日唯一のものであるがこれは進行度分類が治療前に決定されることが必要である以上止むを得ない。しかしこの臨床的分類が実際の癌蔓延の状態と必ずしも一致しないことは当然である。これについてはわれわれも剔出物の詳細な組織学的所見からしばしば報告してきたが、今回は頸癌 80 例の剔出物について組織学的所見を基にして癌進行度の組織学的分類を試みたので報告する。まず子宮を U、陰壁を V、旁結合織を P、骨盤内リンパ節を L とし、癌腫発育の量的状態に主眼をおいてこの 4 部位における癌病変の拡りを観察した。すなわち子宮 U で癌が子宮陰部に限局するを U_F 、頸管に限局するを U_C 、混合型を U_{CF} とし、陰壁 V で蔓延陰性を V_0 、陽性を V_1 、また旁結合織では蔓延陰性を P_0 、子宮側蔓延を P_1 、中央部蔓延を P_2 、骨盤側蔓延を P_3 とする。なおその蔓延様式は問はない。さらに骨盤内リンパ節転移陰性を L_0 、陽性を L_1 とし、これらを総合して 4 級に分類した。I 級は癌病変が子宮 U のみ、すなわち $U_F V_0 P_0 L_0$ 、 $U_C V_0 P_0 L_0$ 、 $U_{CF} V_0 P_0 L_0$ である。II 級は U の他は $V_1 P_1 L_0$ で V_1 、 P_1 の 1 つでも陽性ならこれに入れる。III 級は U の他は $V_1 P_2 L_1$ で P_2 、 L_1 の 1 つでも陽性ならこれに入れる。IV 級は U の他は $V_1 P_3 L_1$ で P_3 なら V_0 、 L_0 でもこれに入れる。

以上の基準にしたがって 80 例を分類するに、I 級 30 例、II 級 19 例、III 級 28 例、IV 級 3 例で、臨床的進行期と一致したのは 30 例 37.5% で 24 例 30% は術前より軽度の、また 26 例 32.5% はより進んだ分類に入れられ、術前診断は進行期を比較的較く評価するのではな

いかと考えられる。

さらに患者予後との関係を 3 年以上経過した 36 例の生存率でみると臨床的進行期では I 期 83.4%、II 期 85.0%、III 期 60.0% とその予後判定は困難であるが、本分類法では I 級 100.0%、II 級 87.5%、III 級 54.5%、IV 級 0% と予後推定上かなり合理的と考えられる。

しかしながらなお本分類法は不完全でありかついくつかの問題があり今後さらに検討の必要がある。

附 議

藤井純一：子宮頸癌の場合、いわゆる臨床的進行期と剔出物を組織学的に検索した組織学的進行期との間に相当の食い違いがあることは当然のことである。われわれも子宮頸癌手術例につき、卵管卵巣、子宮体

部、旁結合織、腔壁への癌蔓延につきしばしば報告して来たが、とくに臨床的癌進期決定の目安となる旁結合織への触診上の所見と、実際の癌蔓延との比較を追加する。進期との関係では旁結合織に触診上の浸潤のないⅠ期(26例)でも42.3%に実際の癌蔓延を認め、Ⅱ期では、53.3%、Ⅲ期で76.9%の癌蔓延を認めている。この旁結合織への癌蔓延と予後(5年治癒)との関係では、癌浸潤なき39例では89.7%の治癒率であるに比し、旁結合織蔓延例では46.7%の5年治癒率である。

原発巣のCPL分類と旁結合織への癌蔓延との関係は、C型で、36.1%、P型で50%、L型で70.5%の蔓延を示している。

齊藤：われわれも旁結合織における組織学的蔓延の高度になるにしたがって患者予後不良の結果を得ております。

115. HISTOPATHOLOGICAL STUDY ON CARCINOMA OF THE UTERINE BODY

GORO IZOE and JUN'ICHI FUJII

(Dept. of Obst. and Gynec., School of Med., Nagasaki Univ.)

Various discussion related to the choice of surgical technic on carcinoma of the uterine body has been carried out by many authors.

For the solution of this problem, the extent of carcinomatous lesions of the uterine body, especially lymph node metastasis was studied histologically in 22 surgical cases, and the following results were obtained:

19 cases out of 22 were adenocarcinoma.

No lymph node metastasis was found histologically in the recent 16 cases which were exstirpated either by simple hysterectomy with lymphadenectomy or by Okabayashi's method, indicating low incidence of lymph node metastasis.

One case of metastasis in the mucous membrane and another in the wall of the Fallopian tube were found. Destruction of the serous membrane of the uterine body was also found in one case.

The carcinomatous infiltration extended beyond the internal orifice of the uterus as demonstrated in 3 cases and extended more than one half of the muscle layer in 6 cases (28.5 per cent), and many carcinomatous embolies were found in the lymph vessels of one case.

8 cases out of 16 were L-Form of CPL classification.

All of recurrent 4 cases died and two of them were C-Form and one was L-Form, thus revealing that in carcinoma of the uterine body C-Form was not necessarily favorable from prognostic viewpoint as compared with that of the uterine cervix.

It would be too hasty to induce conclusion from these small number of cases, but

considering the fact that these 2 cases of C-Form showed the extension of lesion into the cervical canal, the prognosis of the carcinoma of the uterine body seemed to depend more upon the extent than the histological grade of malignancy, thus requiring our further study on this problem.

In one recurrent case autopsy revealed metastasis in the lung, liver and other organs.

附 議

鈴木忠雄：われわれの自験手術例 92 例の転移所見について追加する。

転移のないもの 75 例 (81.5%), 転移のあるもの 17 例 (18.5%) であって、転移症例のうち 8 例 (47%) は付属器転移であり、他に腹腹、リンパ節、旁結合組織の転移が各 2 例、腔、肺、脳、骨の転移が各 1 例である。

転移のない例の 68.0% は腫瘍が限局型で、浸潤性発育型は 32% であり、転移のある例では 17.6% だけが限局型、82.4% が浸潤型であった。

筋層内浸潤についてみるに、転移のない例では、内膜のみもしくは筋層 1/3 までしか侵されていなかったものが 69.3% を占めるに反し、転移のある症例では 64.7% が筋層 1/3 以上を侵している。

頭管蔓延は転移のないものでは 8.0%, 転移のあるものでは 47.1% であった。

体癌の手術方法を考える場合、高位の転移が多いこと、および poor risk の患者が多いことの 2 点から、広汎性術式を全面的に採用しても、必しも顕著な治癒率の改善がみられるとは考えられないが、選択的な症例には概手術が用いられるべきであろう。

116. ON THE FINE STRUCTURES OF ADENOACANTHOMA OF THE HUMAN ENDOMETRIUM

TSUTOMU KASUGA and KUNIO OOTA

(Department of Pathology, Cancer Institute.)

Freshly biopsied fragments from cases of human endometrial carcinoma were examined by electron-microscopy of which three revealed findings interpreted as representing adenoacanthoma of the endometrium.

Correlative histologic study of hysterectomized materials were consistent with the interpretation. As controls many neoplastic and non-neoplastic conditions of the endometrium as well endocervix were examined in parallel. The glandular portions of adenoacanthomas were fundamentally consistent with the ordinary adenocarcinoma of the endometrium. The free margins of the carcinoma cells revealed multiple fine villi while they lacked any trace of ciliary structures. The endoplasmic reticula were dilated with relatively electron-dense homogenous material within them.

In the acanthomatous portions the cell borders between two adjoining cells were remarkably smooth unlike in the case of the squamous epithelium and of the ordinary

squamous cell carcinoma. The desmosomes were very well developed. Tonofibrils were seen running in tangential direction to the nuclear membrane and terminating in the dense structures of the desmosomes. The fibrils were small in number and much more delicate as compared with those in the squamous epithelium. The endoplasmic reticula were scanty in number and not dilated.

Coexistence in a single cancer cell of the villi on the free border, tonofibrils in the cytoplasm and dilated endoplasmic reticula was observed in two cases in which cytologic differentiation was high in optical level.

Thus, adenoacanthoma of the endometrium revealed combined presence of two kinds of cytoplasmic differentiation in different directions in a single clone of tumor cells. It is of interest that the same two-way differentiation can coexist even in a single cell. An investigation of the determinant factors in the cytological differentiation of cancer cell will reveal much more in the future.

117. HISTOPATHOLOGICAL STUDIES ON PRIMARY MALIGNANT TUMORS OF THE ADRENAL CORTEX AND MEDULLA

NOBUAKI SASANO and HARUKI WAKASA

(Dept. of Path., School of Med., Tohoku Univ.)

Malignant tumors of the adrenals possess a wide variety of histologic pictures, where as the appearance of either giant cells or small cells can be initiated from the both origin of the cortex and the medulla. The determination of the histogenesis of a tumor was easy sometimes through their histologic characteristics, while it needed extensive research on the tumor in some cases.

Table 1: Classification of the 13 cases of malignant tumors of the adrenals

Cortical Carcinoma	4 Cases
Giant-cell Carcinoma	2
Small-cell Carcinoma	1
Clear-cell Carcinoma	0
Papillary Carcinoma	1
Tumor of the Medulla	9 Cases
Sympathogonioma	5
Sympathoblastoma	1
Malignant Pheochromocytoma (Pheochromoblastoma)	3

The authors classified the tumors of 13 autopsy cases as shown in the Table 1. In addition, 4 cases of extra-adrenal neuroblastoma and 2 of alveolar soft part sarcoma were also studied considering the histological identification of tumor of the medulla.

Malignant pheochromocytoma with typical endocrinological disorders found in a 24-year-old man, showed pleomorphic picture containing giant cells and spindle cells. These characteristic features were observed either in the both adrenals or in the small metastatic lesions in the thyroid and the liver.

Pheochromoblastoma contained sometimes giant cells with bizarre nuclei, in which the chrom reaction was negative and it was sometimes quite difficult to differentiate this from the cortical carcinoma with giant cells. The former, however, retained the structure of alveolar soft part sarcoma in some parts, and never had diffuse coagulation necrosis besides hemorrhagic softening. While in the cortical carcinoma, character of the cortical cell-cords and/or solid alveolar arrangement of the cells was usually found in some parts of the tissue. Small-cell carcinoma was extremely cellular resembling neuroblastoma but lacking rosette formation, and transition from the dark or light cells retaining the cortical character was sometimes observed.

Characteristic papillary carcinoma which was a head-size tumor of the right adrenal, was found in a 15-year-old boy with clinical manifestation of pubertas precox and the metastases were observed in the lymph-nodes, the liver and other organs.

(人癌の発生と成長 文部省総合研究)

附 議

牛島 宥：1. 副腎皮質癌は比較的小児に多く、また皮質由来を示す各種の移行像を呈するものが見られる。一般に皮質癌の組織像は variation が広いので部分像では分類はむづかしいが示された例は年齢的にも像でも基本像よりはなれたものと思われる。小児例も加え全像としての像の variation を把握しその基準のもとに分類鑑別の問題を処理するが希ましい。

2. 貴症例中の Adrenogenital syndrome を呈した例で細胞的にそれらを基礎づけるようなものが認められたか？

笹野：Adrenogenital syndrome を伴った例は通常認められる大型細胞からなるものではなく、むしろ腎癌や睾丸癌に近いものであった。

おっしゃるとおりの小児癌は手術例などではみているが、剖検例が手許にないので表に入れなかった。表の中で giant cell carcinoma としたのが polymorphzelliges carcinoma に相当する。

118. HISTOLOGICAL CLASSIFICATION OF 414 CASES OF BRONCHOGENIC CARCINOMA AUTOPSIED DURING TWO YEARS OF 1958 AND 1959.

TORU MIYAJI, KENZO ISHIDA, NOBUAKI KAMBARA,
RYUHEI TATEISHI, KOJI WATANABE, MOTOYOSHI SASAKI,
YASUO MARUO, MAKOTO MIYAMOTO, SHOICHI YUI
and SHINTARO ISHIHAMA (Dept. of Path., Med. School Osaka Univ.)

A survey on the bronchogenic carcinoma was made on the autopsy cases performed at 32 medical institutions during two years of 1958 and 1959, and 228 cases out of 3151 male, 55 out of 2016 female autopsies in the year of 1958, and 200 cases out of 2927 male and 45 out of 1873 female autopsies were noted. 414 cases of those were histologically studied and classified as shown in the table.

		male	female	sex unknown	Total
Epidermoid carcinoma 122	cornified	67	5		72
	uncornified	14	6		20
	under-diff.	20	4		24
	pleomorphic	6	0		6
Adenocarcinoma 193	differentiated	64	33		97
	under-diff.	73	19		92
	pleomorphic	3	1		4
Undifferentiated carcinoma 80	oat cell	34	9	1	44
	small cell	11	2	1	14
	large cell	10	1		11
	spindle cell	6	0		6
	pleomorphic	3	2		5
Mixed 19	epid.+adeno.	10	4		14
	epid.+undiff.	3	2		5
Total		324	88	2	414

Histology of bronchogenic carcinoma did not show any correlation with age. Concerning metastasis, however, adenocarcinoma showed the highest incidence of metastasis and followed by undifferentiated and epidermoid carcinoma. The result of present classification was compared with two previous classifications carried out by the senior author, one dealing with 381 cases autopsied before 1952 and the other dealing with 357 cases autopsied during 5 years from 1953 to 1957. A remarkable predominance of adenocarcinoma as a common feature through these three classifications was noted.

附 議

滝沢次郎：退形成の強い種類の癌を未分化癌ということに疑義があります。すなわち扁平上皮癌や腺癌の中にも退形成すなわちいわゆる未分化のものがあるわけでありまして、いわゆる未分化癌でも紡錘形の細胞にまで発達しているものもありますのであります。また良性腫瘍でも未分化腺腫というものもあり得るわけですから未分化ということが癌に特有のものでもありません。

次に角化を分類の中に入れることに疑義があります。角化の中には正常の細胞の角化と Parakeratose とありますので、むしろ扁平上皮組織が基底細胞、多形細胞、扁平細胞と進展する状態の方が分類の根本的性格ではないかと考えます。

また粘液形成についても粘液を形成することは上皮細胞の一つの機能を表わしておるにすぎないのでありますから粘液形成のない時にはその分類は慎重でなければならないと思います。

岡野錦弥：一般的な提案として述べたい。私はかねがね腫瘍組織学的表現に未分化という言葉を用いられることについて、胎生学的な未分化状態に Anaplasie を来したという古典的な概念を現在の生物学の立場から再検討すべきであると考えている。また分化と成熟は異なった概念であって、成熟という事実は胎生期にもあるし、また成熟後も例えば白血球や表皮細胞層にもみられることであるのでその点の区別も望ましいと思う。

なお粘液産生や角質形成は機能的な成熟を表現するものと考えています。

田内 久：肺癌における表皮癌あるいは腺癌の言葉の有する内容は、例えば食道ないし皮膚からの表皮癌および腸などからの腺癌という言葉の有する内容とは必ずしもまったく同日に考えることはできないとも考えられます。それはおのおの発生母組織の生物学的性格の差すなわち化生の問題などが重要な要約を形成しています。

また、手術例と剖検例では癌組織像の様相に種々の差がみられる点は私どもも数年来注目しているところですがこれには癌発育過程における組織像の変換が問題になります。ことに非腫瘍性増殖の場合に重化生の現象をみとめる気管支上皮を母組織とする肺癌ではとくにこの点が問題となると思います。しかし一方肺癌におけるいわゆる表皮癌像が腺癌様構造(私のいわゆる非定型的腺癌)を示すことはしばしばであるがいわゆる定型的腺癌とは区別すべきであります。また一方粘液形成像も経過にしたがってみられるようになることもその発生母組織の性格によって説明し得ると思われまう。

宮地 徹：滝沢教授にお答え。

御指摘のごとく、食道では角化していないのが分化でありましようが、癌としての形態学的考察では、角化するということが扁平上皮癌の分化の極限であろう考えます。しかし、角化しているものと非角化のものとのどんな生物学的な差があるか、後者の方が良性ではないかということについては、例数が不足で(すくなくとも 100 例以上の単位が望ましい)まだ分析するにいたっていません。

粘液を分泌するということは、細胞としては機能的分化の進んだものと考えます。しかし、それだけで腺癌とするのではなくて、このような分類では形態学的な所見がもっとも重要であると考えています。

岡野教授にお答え

御意見ありがとうございます。ここで未分化癌というのは、扁平上皮癌および腺癌は癌としては分化したものであるという点から、それらに属せしめることのできないものをさしたので、胎生学的な意味をふくませておりません。退形成癌とよんでもよいと思っています。しかし、未分化癌にいられた燕麦細胞癌の一部に突如として扁平上皮様構造をみる場合がありますが、これなどは未分化癌が分化しうること示しているのではないかと考えております。

田内教授にお答え。

臓器によって同じ名称でよばれていても形態が異なるというのは当然と考えます。例えば、子宮頸部癌の多くは扁平上皮癌とよばれていますが、皮膚のそれとは随分異なり、太田教授によって検討されているのは周知の通りです。

119. ELECTRON-MICROSCOPIC STUDIES ON MAXILLARY CARCINOMA AND NORMAL AND CHRONIC INFLAMMATORY EPITHELIUM OF THE MAXILLARY SINUSES, ESPECIALLY ON ITS SQUAMOUS CELL METAPLASIA

KEISUKE MATSUI, SHOSUKE MORIWAKI and TEIJI YAMASHIRO

(Dept. of Path., School of Med., Tottori Univ.)

Two cases of squamous cell carcinoma of the maxilla, three cases of normal and twelve cases of chronic inflammatory epithelium of the maxillary sinuses were examined histo-pathologically and electron-microscopically.

Results obtained were as follows:

1. The cilia and microvilli of the ciliated cells of the normal maxillary sinuses were regularly arranged electron-microscopically. Non-ciliated cells of the lower layer contained small amount of tonofilaments which measured 80 to 100 Å in diameter besides mitochondrion and submicroscopic granules.

2. In chronic inflammatory epithelial cells of the maxillary sinuses, marked increase in the tonofilaments and tonofibrils which were consist of increased tonofilaments was observed.

3. In the carcinomatous cells, the endoplasmic reticulums and mitochondria were small in number, and they reduced in number untill they disappeared at last as hornification developed. As hornification increased kerato-hyalin and keratin deposited in the area where remarkably increased tonofibrils were seen. Nuclei and the other ground substanses of the cells degenerated markedly and the other organella disappeared in these cells.

120. ELECTRONMICROSCOPIC STUDIES ON HUMAN CANCER

(I) SQUAMOUS CELL CARCINOMA

MASATOSHI SEKI and KIYOSHI TERAOKA

(Dept. of Path. School of Med., Chiba Univ.)

The human squamous cell carcinoma have been compared with normal human squamous epithelium by electron-microscopy. The tissues of esophagus carcinoma and the adult human skin obtained from surgery, were fixed in 1% osmic acid solution (pH 7.4) immediately.

Cancer cells in the peripheral area of the cancer cell nests revealed the appearance resembling that of the cells of stratum spinosum. They had numerous digital cytoplasmic projections on the cell limit, and desmosomes which were rather irregularly shaped than those of the epidermis were observed obviously. Intercellular lumen dilated than those of the epidermis. RNA granules were abundant and clustered as a rosette-like pattern. Mitochondria of the cancer cells were numerous, enlarged, and empty and had irregularly shaped cristae. It was significant that the granules with high electron density were observed here and there in the nucleus. There was the layer of reticular fibers surrounding the cancer cell nests and the narrow clear space was observed between the basal cytomembrane and the layer. But sometimes, the layer of reticular fibers was absent partially and the cancer cells were exposed to the interstitial lymph spaces. The closer the cancer cells were situated to the center of the nests, the more increased the numbers of tonofibrils and microbodies within the cytoplasm. The condition was reverse as regards the mitochondria and endoplasmic reticulum. The hornification mechanism was just like the modification of that of normal epidermis. But the cells which were in the central part of the cancer cell nests and corresponded to the cells of the stratum granulosum of the normal epidermis revealed enormously large size and arranged as onion skin.

It might be rather rare that the cancer cell nests possessed such regular arrangement of the cells as mentioned above. Significant differences of the cytoplasmic electron density, of the structures on the microorganellae, and of the developmental grades of the hornification were observed between individual cancer cells. These irregularities or "atypism" revealed by electron-microscopic studies might be one of the significant characters of the cancer cells.

附 議

柏井浩三：一般に癌細胞内の糸粒体については、大きさの縮小および数の減少がみられるという報告が多い。先生によれば、ともに、この逆の知見を得られているがこの点に関してどのように考えておられるか。

関：ミトコンドリアの数は、腫瘍の種類によってまちまちな報告があります。私はそれよりもむしろ、Cristaeの不規則な事、Matrixの少ない事を重視したいと考えます。

松井敬介：癌細胞には小胞体やPalade顆粒、あるいは糸粒体が増加することであるが、われわれが観察した前立腺癌では左様であったが（昨年会で発表）演 119 で示したように、上顎癌では減少していた。文献的にも癌細胞にこれらの減少を認めているものが少なくない。

一般論としてはPaladeもいっているように、これらOrganelleの多いことは発育旺盛な細胞の特徴だといえようが、癌のばあいには、発生母組織の種類あるいは癌の種類によって増加しているものと減少しているものがあるのでOrganelleの増加が直ちに癌細胞の特徴とはいえないと思う。

関：ミトコンドリアに関する御意見にはまったく同感であります。このような小器官の数、形態分布などが個々の腫瘍、ないし個々の細胞において不規則であることが、癌の特徴の一つであると考えられます。

121. ELECTRON MICROSCOPIC STUDIES ON CHORIONEPITHELIOMA

YORISATO MORI, MASAYOSHI HASHIMOTO, AKIRA KOMORI,
MITSUO KOSAKA, TOSHIO SHIMOYAMA and KATSUhide AKASHI

(Dept. of Obst. and Gyne., Sapporo Med. College)

The authors have previously reported on electron microscopic studies on hydatidiform mole. In the present study observations on ultra thin sections of tumor tissue with a histological picture of chorioadenoma destruens and choriocarcinoma were made. Whereas, electronmicroscopically, common finding between both tumor tissues were irregular aggregation of the nuclear substance and enlargement of the nucleolus and nuclear pore, these were already known to be common characteristics of tumor cells. The free surface of the syncytium cell seen in chorioadenoma showed a remarkable in-curve and out-curve at times and showed complicated fusion of rod shaped or polypoid shaped microvilli. In addition, near the free surface fine visicles with a fragment opening of the same were seen. The cytoplasm was filled with vacuolar endoplasmic reticulum, while in the cytoplasmic matrix Golgi field and small mitochondria with irregular cristae were seen. Likewise, numerous round or drop-like shaped lipid granules and oval shaped granules of medium density measuring up to 1.5 μ were present. Langhans' cell showed no remarkable differences from that in normal placenta. The microvilli of the syncytium cells in choriocarcinoma were small and rod shaped, while the endoplasmic reticulum were vacuolar and most of the mitochondria showed swelling. There was a scattering of drop-like shaped lipid granules in the cytoplasm. The majority of the cell membranes of Langhans' cells showed remarkable infolding. In most cases the endoplasmic reticulum were observed in parallel with membranes forming narrow spaces and vacuolar shaped endoplasmic reticulum were scarce. The cytoplasm contained drop-like lipid granules around mitochondria and moderately developed Golgi fields. Frequently, the cytoplasm adjacent to the nucleus increased in density by an accumulation of fine fibrous component in the ground substance, in which a number of dense oval shaped particles with doughnut-like central light area approximately 230-270 m μ in size, were seen.

122. ELECTRON-MICROSCOPIC STUDIES ON CLEAR CELL CARCINOMA OF THE KIDNEY

TAKAMITSU KUSUNOKI, KOOZOO KASHIWAI, HISAO YANO
and TAKEZOO MATSUNAGA

(Dept. of Urology, Med. School, Osaka Univ.)

The fine structure of clear cell carcinoma of the kidney was examined by electron microscope and compared with that of human fetal adrenal cortex as well as that of the fetal kidney.

1. Clear cell carcinoma of the kidney: The cells filled with fine granules of low electron density were predominant and they were mixed with dark cells having homogenous cytoplasmic matrix of comparatively high electron density. Cytoplasm had extremely variable in both light and dark cells. They often contained lipid droplets and vacuoles of highly variable size and shape.

In some place, cytoplasm showed marked vacuolisation.

The mitochondria also presented various morphological appearance. It was interesting to note that some mitochondria contained tubular or villous structure, which was usually observed in adrenal cortex.

In intercellular space, there were microvilli differently developed, and remarkably developed one simulated brush border of renal tubular cell.

In some place, there were subendothelial spaces being adjacent to pseudosinusoids. Such structure was similar to sinusoid of adrenal cortex.

2. Inner zone of fetal adrenal cortex: There existed also both light and dark cells, and their cytoplasm contained variable lipid droplets and vacuoles.

There were also cells in which cytoplasm showed marked vacuolisation.

Microvilli were detected in intercellular space too.

3. Fetal renal tubules: The brush border was similar to remarkably developed microvilli of clear cell carcinoma of the kidney.

Other similarity between the tumor cells and fetal renal tubular cells could not be found.

123. ELECTRON-MICROSCOPIC OBSERVATIONS ON SARCOMA INDUCED IN THE SUBCUTANEOUS TISSUE OF THE RAT BY ADMINISTRATION OF TRYPAN BLUE

TEIICHI TESHIMA, KATSUHIRO NAKAMURA and TAKUZO ISHIDATE

(Dept. of Path., School of Med., Tohoku Univ.,)

21 cases of sarcoma which were induced in the subcutaneous tissue of the back of Wistar-strain rats were examined by electron-microscope.

1 cc. of 1% saline solution of trypan blue was injected into the subcutaneous tissue of the back of 130 rats once two weeks. 53 of 130 rats which survived seven months had received 13 injections. The other 77 rats were sacrificed periodically as control. Trypan blue injection was continued on 53 rats eight more months and 1 to 6 injections were added on each rat by the end of experiment. Tumor growth was first noted at the injected site seventh months after injection and 21 of 53 rats (39.6%) revealed sarcoma formation.

Histologically, they were divided into 2 types. One was 15 cases of fibroplastic spindle cell sarcoma, two of which showed remarkable fiber formation, and the other was 6 cases of fibroplastic polymorphic cell sarcoma.

On electron microscope, cells of the tumor tissue were classified into two groups according to the type of endoplasmic reticulum. Group I consisted of cells in which rough surfaced endoplasmic reticulum was prevalent. This group was again divided into two according to the shape of rough surfaced endoplasmic reticulum; immature cells with short, tubular endoplasmic reticulum and mature cells with extended reticulum, filled sometimes with the homogeneous substance of medium density. Group II consisted of cells in which smooth surfaced endoplasmic reticulum was prevalent.

Electron microscopic findings on the fibers in tumor tissue were as follows:

- 1) The fibers measured approximately 200 to 700 Å in diameter, and became thick bundle in two cases with remarkable fiber formation.
- 2) Almost all the fibers had no cross striation.
- 3) Fibers were observed only extracellularly.
- 4) Some fibers were found in the amorphous substance of medium density.
- 5) There was no definite findings which suggested that only cells containing rough surfaced endoplasmic reticulum filled with homogeneous substance could produce fibers.

124. ON TUMOR OF DOGS

TANETAKA CHIBA (Reserch Institute of Environmental Medicine, Nagoya Univ.)

Case 1. Epithelioma Adenoides Cysticum.

A male shephard dog. (5 years of age).

In June 1960, many small size nodules of all parts, especially at neck of skins were observed.

Biopsy. The tumor were solid beneath the cutis. Some of them showed on the cutis as cabbage-like lumps of lipoid or keratoid substance.

Histologically, the tumor consisted of aggregated epithelial cells which were pressed inside through the cutis into the corium, where they prolifereted. They were arranged spherically at many sites, with a strongly confined area in the center. This case was the same as Sakurai's report. (Jour. Jap. Vet. Med. Assn. 1960. No. 5)

Case 2. Reticulosarcoma.

A male mongrel dog. (12 years of age).

In the Oct. 1958 a swelling of the fore-head was noticed and the animal seemed to be in malaise. The swelling was found to have spread to the nasal-cavity and eyelid from fore-head.

A biopsy of the swelling of the fore-head revealed a reticulosarcoma. Treatment by the brun of tumor tissue was attempted, but the animal died on Sep. 2. 1960.

The nasal-cavity was occupied by a neoplastic mass and it showed a uniform, greyish-white, soft substance. Microscopically the tumor consisted of a mass of reticulum cells.

125. HISTOLOGICAL AND HISTOCHEMICAL STUDIES ON THE DUCT-LIGATED SALIVARY GLANDS IN RATS

AKITOSHI SUGIMOTO (Dept. of Oral Path., Dental School, Osake Univ.)

The secretory ducts of submaxillary and sublingual glands of the right side were ligated respectively, while the left glands remained always as controls. Three major salivary glands were observed histologically and histochemically (for alkaline and acid phosphatases, esterase, polysaccharides, nucleic acids, protein-bound amino- and SH.SS-groups) 1 to 7 and 14 days after ligation. The results obtained were summarized as follows: (1) The ligated glands increased in weight with swelling,

hyperemia and edema during first 5 days. Then the glands fell into marked atrophy, showing turbidity and hardening. (2) Histologically, the ligated submaxillary glands showed a marked necrosis, and then a remarkable regenerative proliferation of the surviving parenchymal cells appeared 3 days after ligation, accompanying granulomatous tissue. Three types of the proliferating cells were seen. Type I seemed to be of the serous acinar cell, proliferating only during first few days. Type II was of the intralobular duct cell, proliferating rapidly during 4 to 6 days, forming an adenoma-like or cyst-like structure with a marked squamous metaplasia and cornification. Type III was of the interlobular duct cell, proliferating slowly but continuously, and showing a slight squamous metaplasia. The duct-ligated sublingual glands also fell into a marked necrosis, having no tendency of proliferation. (3) Histochemically, the proliferating cells of each type showed some characteristic reactions. Type I possessed a highly increased reaction for alkaline phosphatase. Type II showed marked increased reactions for phosphatases, esterase, amino-group, and especially for PAS and SH-SS groups in the cornified metaplastic areas. Type III showed PAS and SH-SS reactions similar to those of Type II, although very slight in degree. (4) From these observations it seemed to be concluded that in regenerative proliferation after the duct-ligation of the submaxillary gland in rats, the ductal elements played a significant role, showing a high squamous metaplasia.

V. Transplantation

126. METABOLISM OF ^{32}P IN EHRlich ASCITES TUMOR CELLS AFTER DESTRUCTION OF THE CELLS (I)

SIGEKAZU MAEDA (Dept. of Obst. & Gyne. Osaka City Univ.)

Ehrlich ascites tumor cells were transplanted into the abdominal cavity of mice. On the third day after inoculation, ^{32}P (100 $\mu\text{c}/\text{animal}$) was injected intraperitoneally. On the 7th day, the ^{32}P -labeled tumor cells (Group I) were obtained and non-cells layer (Group II) was separated by centrifuging. Cell-membrances of ^{32}P -labeled tumor cells were washed with NaCl solution and destructed by homogenization as completely as possible. (Group III). An equal volume of 1N HCl was added to the destructed tumor cells, and this mixture was homogenized again and was washed (Group IV). The precipitate of each group was mixed to non-labeled tumor cells. The mixtures were transplanted into the peritoneal cavity. The distribution of ^{32}P in several organs was measured by G-M tube on the 7th day after inoculation.

RESULTS

Group I: C.p.m. of ascites cells was higher than c.p.m. of other organs. Uptake of ^{32}P of bone in group I showed moderate grade as comparing with that of other groups.

Group II: No remarkable ^{32}P -incorporation into Ehrlich ascites tumor cells was recognized. But uptake of ^{32}P of bone and muscle was higher than that of other organs relatively.

Group III: The increase in ^{32}P -uptake of bone was seen, but ^{32}P -uptake of liver and muscle was decreased.

Group IV: The increase in uptake of liver, bone and spleen was relatively shown. It may be concluded that ^{32}P in ascites tumor cells may be utilized to metabolic system of normal organs again.

127. EXPERIMENTAL STUDY OF HUMAN CANCER CELL TRANSPLANTATION INTO THE GERM-FREE ANIMALS USING MILLIPORE FILTER (II)

MASASUMI MIYAKAWA, SOICHI IJIMA, HIDEMASA KISHIMOTO,
YUTAKA UNO and HIROSHI MIDORIKAWA
(Dept. of Path., School of Med., Nagoya Univ.)

The authors traced the fate of human cancer cells grafted in the diffusion chamber in the abdominal cavity of the experimental animals. The chamber consisted of a cylinder some 5 mm. in diameter and 1 mm. deep. The top and bottom of the cylinder were closed by discs of millipore filter sealed with acriloid cement. The millipore filter made barrier between the tissue of host animals and grafts, providing the invasion by various kind of wandering cells. Nutrient and other substances could pass into or out of the chamber, and host cells could be either admitted or excluded by varying the pore size of the diffusion membrane. The DA millipore filter of average pore size 0.65μ was used for most of the experiment.

Ascites tumor cells of rat, Yoshida sarcoma, survived and proliferated for long period of time about 25 days in the cell-impermeable chamber in the abdomen of rat. In the experiment using guinea pigs, ascites tumor cells of rat grew progressively in the chamber for 10~12 days after a sharp, early drop in cell number, and then were destroyed.

The human cancer tissues (epidermoid cancer), transplanted in the subcutaneous tissues of the back of the experimental guinea pigs, were early infiltrated by host cells and underwent necrosis after 4 days. On the other hand in the diffusion chamber the grafts survived for 10 days, and then fell into necrosis.

These shows that cell fixed antibody plays a roll at first and then circulating antibody plays an important roll in the tumor-heterotransplantation.

附 議

秋山武久: 1. conventional な抗体の filter 通過性について基礎的の検討があればお教え願いたい。

2. chamber 挿入にともなう異物反応としての主体側の例えば大網による包蔽作用が体液の chamber 内への透過を強く阻止することはないか。

3. あらかじめ免疫した動物に chamber を挿入して実験したことはないか。

滝沢延次郎: 吉田肉腫細胞を chamber に入れて生存増殖しておる場合と人癌を chamber に入れて行った場合とで chamber の膜の外に集る細胞の種類ならびにその程度について差がありますか。

宇野: Millipore 膜の外側には腹腔内の遊離細胞がひっつかかってきますが、おもに単球好中球等で、吉田肉腫を可移植系動物に移植した場合と、人癌を用いた場合とでは、ひっつかかる細胞はその初期では余り差がないように思われます。Diffusion chamber を腹腔内に容れますと、大網でかるく取巻かれます。

Diffusion chamber 内の液を電気泳動で調べた実験は目下検討中でありまして、はっきりしましたらのち程報告いたします。あらかじめ免疫しておいて行う実験は今回は行いませんでした。

128. ĈELOSTAMO DE MUSA MAMA TUMORO (SATŌ-KANCERO N-RO 1) EN KONTINUA KULTURO *IN VITRO*

YOSHIHIRO YASUMURA kaj TSUGUO KUWATA

(Bakteriologia Instituto, Medicina Fakultato, Tiba Universitato)

Nuntempe virusologoj kaj onkologoj uzas establitaĵajn ĉeloliniojn precipe por analizi, sub la ĉela nivelo, sian temon pri gastiganto-parazita rilateco kaj pri onkogeneco. Tia afero ĉiam postulas, ke al la listo, kiel eble pli da novaj ĉelolinioj devas esti aldonataj, kiuj ludus novan rolon en esplorkampo. Tiu ĉi raporto koncernas: a) devenon kaj disvolvon de kontinua ĉelolinio de musa tumor, kaj b) citopatogenan efekton (C.P.E.) de polioma viruso en tiuj ĉi celoj *in vitro*. Metodo estas esence tia sama, kiel antaŭe ĉe la stablo de la ĉelolinio "FRUKTO" de musa Fruktoza sarkomo (GANN, 50 (Suppl.) 182, 1959). Tumora materialo estis tripsinizata laŭ la metodo de RAPPAPORT. Proksimume 10^6 da celoj/ml estis semataj en boteletojn, poste inkubatajn en 37°C , po 3 ml en nutra medio (PARKER'a 199-solvaĵo kaj ĉevala sero 20%). Ekkomence celoj difuze algluiĝis al vitrosurfaco kaj tamen ĉ. 1-2 semajnojn poste grandparte degeneris. Post unu monato, neatendite ekaperis en du boteletoj kelkaj vivecaj ĉelokolonioj (diametro ĉ. 1-2 mm) kiuj tamen kreskadis malrapide. Intertempe, ĉe unu boteleto, post 5 monatoj, kiam la kelkaj kolonioj kreskis je ĉ. 10 mm diametre, malsukcesa subkulturo estis provata, uzante tripsinon. Ĉe la alia boteleto, kiu naskis nunan ĉelolinion "SAKO" kaj tiam enhavis ĉ. 2.5×3.5 cm-an kolonion, subkulturo sukcesis finfine post 10.5 monatoj ekde la komenco, kvankam unua pasigo ĉe la sama boteleto je 6 monatoj ne fruktodoniĝis tiam, kiam aliaj kolonioj estis, gumospatele skrapite subkulturitaj. Poste celoj bone kreskadis kaj ĝis nun trapasis 15 subkulturojn dum 15 monatoj. Pasiĝo okazas intertempe de meznombro 8 tagoj. Hodiaŭ estas ankoraŭ havebla unu varianta-linio, adaptita al medio, YLE-solvaĵo 90%: Bovida sero 10%. La originala kaj varianta celoj havas preskaŭ saman aspekton epitelisimilan. Ili ambaŭ subtenas kreskon de polioma viruso, kiu donas klaran citopatan efekton al la celoj.

**129. MILKY SECRETION IN A TRANSPLANTED MAMMARY CANCER
OF THE DD MOUSE (I) INFLUENCE OF SEX HORMONES ON
THE SECRETORY ACTIVITY AND TRANSPLANTABILITY**

HIDEO MIYAWAKI and SHOZO ISHII**

(Dept. of Path., School of Med., Mie Prefectural Univ.)

(1st Surgical Division, Med. School, Kyoto Univ.**)

A reddish-brown tumor originated spontaneously in the right second mammary gland of a 11-month-old female dd mouse. Serial transplantation of the tumor was made into the brains of female dd mice up to the 12th generation. After the 11th generation, the tumor was maintained also by serial subcutaneous transfer until the 15th generation. Due to lack of strain prototype of the dd mice, only 70 per cent of 228 mice developed tumors during the period of 1 to 2 months after transfer. It was revealed that transplantation to the male mice was always unsuccessful, or otherwise, the growth of transplanted tumor was markedly retarded. Histologically, the original tumor was an adenocarcinoma with large blood-filled cysts in the sheets of small acini. If any secretion was noticed in the lumens of acini, it was scanty and was made up of homogeneous, eosinophilic material which indicated the positive PAS reaction. The histologic appearance of the transplanted tumors remained unchanged during the first three generations. After the 4th generation, however, variation to fleshy white tumors occurred and some of them showed conspicuous milky secretion. Histologic examination revealed abundant sudanophilic fat globules, aggregating in a finely granulated, PAS-positive, albuminous fluid. Secretion was pronounced exclusively in the case of brain transfer.

No tumors developed among the groups of spayed female or non-treated male mice, transplanted subcutaneously with the tumor during observation for 90 days (0/6 and 0/5, respectively). On the other hand, tumor takes occurred in three groups of spayed female mice treated with 15 injections of sex hormones as follows: estradiol 4 γ , progesterone 0.1 mg or 0.5 mg, every other day, respectively (3/5, 3/5 and 2/5). The same dose of estradiol was also effective for obtaining adequate tumor growth in the male mice. The transplanted tumor had a distinct response with patchy lactational changes, to estradiol or progesterone.

It was concluded that the tumor concerned was biologically hormone-dependent and functionally hormone-reactive.

130. MILKY SECRETION IN A TRANSPLANTED MAMMARY CANCER OF THE DD MOUSE (II) HISTOCHEMICAL STUDIES OF LIPIDS

HIDEO MIYAWAKI and SHOZO ISHII*

(Dept. of Path., School of Med., Mie Prefectural Univ.)

(1st Surgical Division, Med. School, Kyoto Univ.*)

Part one of this paper reported that milky secretion appeared in a mammary cancer of the dd mouse during the course of serial transfer. In the present work, comparison was made using a variety of histochemical methods for lipids between the milk of normal lactating mammary glands and the milk-like secretion of transplanted mammary cancers of the dd mice.

The alveoli of the normal mammary glands at their fully established stage of lactation were distended with milk which contained numerous sudanophilic fat globules in the PAS-, HgBPB-positive, albuminous fluid. These fat globules exhibited positive reactions for neutral fat, phospholipid, and fatty acid. On the other hand, the transplanted mammary cancer secreted two kinds of globules together with the finely granulated albuminous fluid which gave positive PAS and bromphenol blue stain. One of these globules was almost identical in histochemical character with the fat droplets in the normal lactating mammary glands, as revealed by positive neutral fat, phospholipid, and fatty acid reactions. The other globules were quite different in morphological and histochemical character. They were spherical or spheroidal, and tended to coalesce each other to form a large bizarre-shaped globule. These globules gave positive reactions for cholesterol and fatty acid. They were insoluble in organic solvents, retained in paraffine sections, stained intensively with bromphenol blue, and gave a negative reaction with PAS. Thus, the latter globules were regarded histochemically as lipoprotein.

The present histochemical observations indicated that function of normal lactating mammary tissue, the production and secretion of milk, was reproduced only partially and incompletely in this particular mammary carcinoma. As to the problems in regard to the elaboration of cholesterol-bound protein by cancerous mammary tissue, no definite conclusion could be drawn in the present histochemical study.

131. AN EFFECT OF ACID MUCOPOLYSACCHARIDE ON THE TRANSPLANTATION AND GROWTH OF TUMOR

JUN TAKEUCHI

(2nd Dept. of Path., School of Med., Nagoya Univ.)

In the previous paper it was shown that a metachromatically colored spot with toluidine blue appeared on the filter paper after electrophoresis of alkaline extract of Brown-Pearce carcinoma tissue and was situated on the place corresponding to that of chondroitin sulfate.

In the present study the spot was stained orange-yellow by acriflavine solution, indicating the existence of the sulfated radical.

In order to investigate the relationship between tumor growth and sulfated acid-mucopolysaccharide, tumor tissues were emulsified with phosphate buffered saline (S group) and with chondroitin sulfate solution (Ch group), and these emulsions were subcutaneously inoculated in each rabbit separately into the abdominal region at the same time. Day by day the tumor growth was examined by measuring two diameters of the tumor masses comparing between these two groups. From 1st to 15th day tumor masses in Ch group were larger than those in S group. One or two days after inoculation the tendency of degeneration and necrosis of the tumor cells in S group was more markedly seen histologically than in Ch group. About five days after inoculation the tumor mass became to be almost occupied by vigorously proliferated tumor cells.

In this case, tumor cells of each group kept in emulsion at room temperature for 30 minutes were stained with Nigrosin (by means of Kaltenbach's method) for differentiating live and dead cells, but there were not recognized any significant differences in the live cell per cent among inoculated tumor cells between two groups.

In the course of 10 hours duration after inoculation, there could be demonstrated, in the tumor tissue fluid, chondroitin sulfate electrophoretically on the filter paper.

There may be considered the existence of some protective actions for the inoculated tumor cells by chondroitin sulfate and this action may be due to negative electric charge of sulfated acid mucopolysaccharide or to viscosity of it.

132. ANTITUMOURAL EFFECT ON TRANSPLANTATION OF ASCITES HEPATOMA AH 130. (I.)

RYUJI MIZUMOTO and AKIRA NODA

(2nd Dept. of Surgery, Med. School, Kanazawa Univ.)

The rats, which showed spontaneous regression after the implantation of weakened cells, at low temperature, of ascites hepatoma, had a tendency to stand against re-implantation of the tumor. Sera of those animals were examined serologically, in order to clarify their tumour-inhibitory effect, employing tumour agglutination test, precipitation test, complement-fixing reaction and tanned cell hemagglutination. However, there could not be found any positive findings concerning the existence of tumour-inhibiting factor. Moreover, we examined the spleens of these animals which were said that they might play the most important role in defence mechanism of organism, and found their extraordinary enlargement. We made an experiment to study the role of the spleen of tumour bearing rats in tumour-inhibiting effect.

Rats of two groups were implanted ascites hepatoma AH 130 intraperitoneally and subcutaneously, respectively. The rats of each group were divided into three groups, (1) control rats, (2) rats which received injection of spleen-homogenate of normal rats, (3) rats which received injection of spleen-homogenate of tumour bearing rats. Spleen-homogenate was injected subcutaneously for three times, 0.5 cc each time.

As to intraperitoneally implanted group, rats treated with the homogenate of tumour bearing ones lived a little longer than two others. Density of tumour cell was also lower, other findings being similar to the control.

As to subcutaneously implanted group, 81.8% of the rats which received injection of the homogenate of tumour bearing rats survived and their tumour showed gradual regression, while 10.1% of control rats and 20.8% of the rats injected the homogenate of normal ones showed the similar course. Results of other examinations went parallel with the course of subcutaneous tumour. Histological finding of the tumour revealed that, as days went on, tumour tissue showed tendency of healing being surrounded by granulation and collagen fiber, and finally disappeared.

We are studying about cross test in the same way between ascites hepatoma and Yoshida sarcoma, in order to clarify, whether this tumour-inhibiting effect is specific or non-specific.

133. SURVIVAL, POLIFERATION AND MORPHOLOGY OF NF SARCOMA TISSUES EXPOSED TO LIQUID NITROGEN

IWAO NAGAI, WATARU YOSHIOKA, YOSHINORI TANAKA
and MASAYOSHI KUMEGAWA

(Dept. of Oral Surgery, Dental School, Osaka Univ.)

The survival and proliferation of mouse frozen NF sarcoma tissues have been quantitatively estimated by determination of the rate of isologous transplantation. The sarcoma tissues were chopped out, pretreated in 30 per cent glycerol, frozen rapidly in liquid nitrogen (-195°C), rewarmed rapidly at 37°C in Hank's solution, and then transplanted to the axillae of adult mice.

The results were checked by morphological and pathological examination of the frozen tissue and by *in vivo* growth.

1) Both glycerol treated control and intact control group of NF sarcoma tissues showed the highest proportion of successful takes, 100 per cent growth in axillae.

2) The tumor tissues that were exposed to glycerol but not frozen showed a few damage compared to intact control tissue and the glycerol solution in itself caused some injury under the conditions of this experiment.

3) Pretreatment with glycerol increased the proportion of successful transplants among frozen sarcoma from 64 to 73 per cent in tissues transplanted to the axilla, however, showed pathologically a little cell damage caused by freezing in the tissue before transplantation.

4) The tissue frozen without pretreatment in glycerol showed almost the same histopathological findings with the group of glycerol treated frozen.

5) On tumor growth, both group of frozen with and without pretreatment in glycerol took more time than intact and glycerol treated control group.

6) If the tumor transplants once grew, no significant difference in histopathological findings was observed among each group.

7) Good correlation was observed between the morphological characteristics of tumor tissue and biostatistical data of growth tumor tissue after *in vivo* transplantation.

附 議

乾 直道：凍結をうけた癌組織が組織像に顕著な変化がないことを伺いましたが組織を形成しているおのこの細胞の性格（染色体、その他細胞内物質の変化）をしらべていられたら御教え願いたい。

吉岡：無処置対照群に比し、凍結同群において示した細胞変性の程度はごくわずかでありますがとくに申しますならば、細胞原形質内に多少の空胞変性があり同時に細胞自体わずかに萎縮しておりました。核の変化は各群間にほとんどその差は見られませんでした。

134. STUDIES ON ASCITIC CONVERSION OF FRUCTOSE SARCOMA IN MICE

TSUGUO KUWATA and HIROSHI MURAYAMA

(Dept. of Bact. & Path., School of Med., Chiba Univ.)

Fructose sarcoma has been maintained serially over 500 hundred generations in mice as a solid tumor and so far examined, non-convertible to ascites form. As UV-irradiation often exhibits mutagenic action on microorganisms, attempts were made to irradiate fructose sarcoma cells *in vitro* and convert it into ascites sarcoma. First, solid tumors were treated with 0.25% trypsin solution and free cell suspension was obtained. The cell suspension was then irradiated with sublethal dose of UV light and transplanted into mice together with normal mouse embryos. Tumors, which subsequently appeared, were extirpated, trypsinized and again irradiated. After these treatment, we obtained a sarcoma cell line which caused metastases in the lung of transplanted mice. This unusual events suggested occurrence of variation in characters of fructose sarcoma. By using this sarcoma subline, ascitic conversion experiments were started. In the first several generations, there were hemorrhagic ascites with few tumor cells. During subsequent passages, volume of ascites and total tumor cell numbers increased gradually. From the 14th generation, ascites became non-hemorrhagic and there were almost no infiltrative propagation of solid tumors in the mesenterium. At the same time, average survival days of ascites-carrying mice prolonged to 20 to 25 days, despite active cell propagation in the peritoneal cavities. Total tumor cell numbers in the peritoneal cavities reached over 10^8 about 10 days after intraperitoneal inoculation. These ascites tumor cell suspensions were titrated through intracerebral, intraperitoneal and subcutaneous routes, and it was found that about 10^5 cells were necessary for the production of ascites tumors. Non-irradiated solid tumors also produced hemorrhagic ascites, but it was not possible to make serial passages. Both lines of tumor cells had almost the same capacities of growth in the brain. Even after intracerebral passages, ascites sarcoma cells were able to grow and form abundant ascites when returned again to the peritoneal cavities.

附 議

滝沢延次郎：果糖肉腫を長く累代移植を行い腹腔内に移植をして見ますと数代位で結節型になる傾向がつかったのですが演者はただいまの方法で腹水型に変えることができましたので腫瘍の研究に新しい領域ができたことになり喜ばしいことと存じます。

私は結節型と腹水型の果糖肉腫細胞に構造上いかなる変化がおきたかに非常に興味を持っております。

川俣順一：われわれは、アクチノマイシン肉腫の発生を 1957 年の本学会で報告したが、本腫瘍の移植継代 11 代目において、腹水型腫瘍の一系統の分離に成功した。現在 100 代以上の継代を行っており、この

腫瘍を用いて、腹水型と腫瘍型腫瘍の相異を検討しているが、移植に当っての系特異性が継代の間に、次第に失われつつある。一方、この腫瘍が制癌実験の際の被験腫瘍として使用できるか否かということについても研究を続行している。

この腫瘍の腹水化は、“gradually”に行われるようで、初期の腹水量は少かった。

桑田：Klein 等によれば固型腫瘍を腹水型に転換する場合、直ちに腹水化されるものと、漸時腹水化されるものとある。われわれの果糖肉腫は後者に属するものと思われるが、アクチノマイシン肉腫の腹水化の場合はいかがでしょうか。

135. STUDIES ON THE SUSCEPTIBILITY OF DDN MICE TO INOCULATION WITH C_{58} LEUCEMIC ASCITES

AKINORI SHIRAISHI (Dept. of Internal Med., Med. School, Okayama Univ.)

An ascites tumor was established in the C_{58} mice by serial intraperitoneal transplantation of the spontaneous lymphosarcoma originated in a female C_{58} mice. 0.1 ml. of this ascites induced the leukemia in C_{58} mice on 7 to 10 days after inoculation. And the same dosis of this ascites inoculated intratesticularly into adult ddN mice produced the nonleukemic ascites tumor with leukemoid blood picture. The ascites growing in C_{58} and adult ddN mice had the property inducing leukemia in new born ddN mice on 10 to 13 days after inoculation. The successive transplantabilities of leukemia in adult C_{58} mice and new born ddN mice and of the ascites tumor in adult male ddN mice were 100% and 70% respectively.

When the cell free filtrate of C_{58} mice leukemic spleen and liver prepared by using the ceramic filter L3 was inoculated into young C_{58} mice, leukemia had developed only in this mice on 28 to 33 days after inoculation. In the first generation, the takes rate of this leukemia in C_{58} mice was 60%, and in following generations the takes rate was turned out to be 100%.

The morphological examination of the leukemia in C_{58} and new born ddN mice, and the ascites tumor in adult ddN mice was carried out. The leukemia developed in C_{58} and new born ddN mice by inoculation of ascites tumor cells was seemed to be myelogenous. But, in the first and following few generations, the kind of leukemia induced by leukemic cell inoculation could be interpreted as the lymphatic, because many immature cells were assumed to be lymphoblasts and there were very few developmental stages. And in following generations, many developmental stages were seemed to be myelogenous appered. Moreover, the peroxydase reaction of peripheral blood smear was found to be positive. And the C_{58} mice leukemia induced by cell free filtrate was also myelogenous.

The ascites tumor cells which induced the leukemia in C_{58} and ddN mice was con-

ceived to be extremely immature blood cells or tumor cells. But the cell free filtrate came from leukemic C_{58} mice could induce myelogenous leukemia in C_{58} mice. Then, it was concluded that the myelogenous leukemia could be induced by a virus like substances.

136. ESTABLISHMENT OF A NEW TRANSPLANTABLE STRAIN OF MURINE LYMPHOCYTIC LEUKEMIA (ASCITES FORM) OHO NO. 1

KOICHI OKADA and NORIO KAWAMURA

(Dept. of Internal Med., Med. School, Okayama Univ.)

By inducing lymphocytic leukemia in DBA mice with 20-methylcholanthrene, it has been converted to ascites form and the transplantation of it is now carried on for 70 consecutive generations.

The survival time of the animal with original leukemia proved to be 29 days, whereas that of the animals transplanted with this ascites was gradually shortened and now it was 8 days. The peripheral leucocyte count in the later stage was $4 \times 10^4/\text{mm}^3$ and the rate of lymphoblast appearance was 20 to 30 per cent.

The volume of ascites reached 1.5 cc, the cell count $27 \times 10^4/\text{mm}^3$, and the protein content 4.6 g/dl. In the observation of ascites cells, the lobulation of nucleus was marked and there were one or two vacuoles in cytoplasm, presenting a weak green fluorescence in the fluoro-chrominized preparations. In counting the number of chromosomes of the 213 ascites cells at metaphase, the majority of the cells contained 30 to 45 chromosomes and the greatest proportion of the cells contained 40, amounting to 26.8 per cent. There were also tetraploid cells amounting to 1.4 per cent. In general the shape of chromosomes was rod-like but some were V-shaped or large rod-shaped.

This ascites could be transplanted even after diluting it down to 10^{-4} time (cell count 3,200) and with increase in dilution the survival time was proportionally lengthened. In preserving the ascites, it was transplantable for 3 days, if kept at 20°C , for one week at 4°C , and for three weeks at -30°C . It was 100 per cent transplantable in the same strain and also transplantable to new borns of the other strains like CBA, B III and C57 black. In the case of mature mice of different strains like CBA and R III intraperitoneal transplantation revealed lethal transplantability and subcutaneous one temporary take. However, no leukemia ensued in transplanting the ascites after repeated freezing-melting or freezing-drying processes. The authors designated it as lymphocytic leukemia OHO (Okayama, Hiraki, Okada) No. 1.

137. FURTHER STUDIES ON THE TRANSPLANTATION OF A MIXTURE OF DIFFERENT STRAINS OF TUMOR CELLS

TORU KUMAMOTO and SEIJI SOEJIMA

(Dept. of Path., School of Med., Hirosaki Univ.)

1) At the last meeting of the Japanese Cancer Association, it was reported that the intraperitoneal transplantation of a mixture of diploid Hirosaki sarcoma and Usubuchi sarcoma in hybrid rats resulted in favor of diploid Hirosaki sarcoma at the 5th day. It was also reported at the same time that the intraperitoneal successive transplantation of a mixture of diploid and tetraploid Hirosaki sarcoma in hybrid rats resulted in favor of diploid Hirosaki sarcoma after about 1 month in the course of 7 successive generations.

2) In the present experiments, successive intraperitoneal transplantations of a mixture of diploid and tetraploid Hirosaki sarcoma in hybrid rats were repeated two times and both resulted in favor of diploid Hirosaki sarcoma as in the previous experiments.

3) In the present experiments, successive intraperitoneal transplantations of a mixture of diploid Hirosaki sarcoma and Hepatoma 7974 in hybrid rats were also carried out and repeated twice. Both experiments resulted in favor of diploid Hirosaki sarcoma after about 2 months in the course of 10 successive generations.

(文部省科学研究費による)

VI. Carcinogenesis

138. CANCER AND MESENCHYMAL TISSUES (II)

MIYOSHI URABE, TETSUJI MIZUKAMI, SHIRO TSUNAMURA,
SEIJI MIYAZAKI, KIICHI WATANABE and TEISUKE TACHIBANA

(Department of Surgery, School of Medicine, University of Kanazawa)

We have previously reported that the mesenchymal tissues had close relation to the growth of cancer. In this report, we are going to present the influence of the several attacks of the peripheral and central nervous systems to the growth of cancer, having direct and indirect effects to the mesenchymal tissues.

In the first place, the ischiadic nerve was cutted and DMBA was applied to the site of the skin where the nerve was cut. It was found that the cutaneous tumor developed remarkably and incidence of the cases accompliced by cancerous changes was also large on the operated side in comparison with the unoperated side. Moreover, it was also recognized that the cutaneous change became worse for the short period by the administration of Cortisone in cases of ischiadic operation.

Secondly, the pallidostriatum in the brain where the huge nucleus is seen has close relation to the metabolism of living body. We have many cancer cases where the regressive degeneration was recognized at autopsy in this area of the brain. And, we could find the significant growth of Yoshida sarcoma after the pallidostriatum of the rats was experimentally destroyed by electrocoagulation or by injection of a solution of corrosive sublimate.

Thirdly, we tried to analyse the so-called "Nekrohormon" caused by the ligation of splenic artery. The extracts produced by salting out method with ammonium sulphate from the spleen, liver and lung after the ligation of splenic artery of the rabbit were injected in rats which were transplanted intrapulmonary with Yoshida sarcoma, and their survival time was examined. As the results, it was confirmed that the extracts activated the mesenchymal tissues more than those of the group where the splenic artery was not ligated.

139. AN AUTOPSY CASE WITH MANY PRIMARY TUMORS

SATOSHI KANO and HISANDO KOBAYASHI

(2nd Dept. of Path., School of Med., Nagoya Univ.)

Here presented an autopsy case of 64-year-old male with many foci of tumorous or hamartomatous proliferation in the organs.

There were no particular pathological findings in his past and family history.

About 4 years before his death, the left kidney (24×11.5×7.5 cm, 1340 g) was sur-

Tumorous or hamartomatous foci will be indicated in the following Table.

Site of the focus	Size and shape of the foci	Histological diagnosis
Upper lobe of the right lung	Nodular 50×30×30 mm	Squamous cell carcinoma from right main bronchus
Metastatic foci of bronchial carcinoma		
Brain		
Middle pons	10×5×3 mm	Squamous cell carcinoma with hemorrhage
Left thalamus	4×5×3 mm	
Right parietal lobe	1.2×0.3×0.3 mm	
Left caudate nucleus	1×2×2 mm	
Right testis	Microscopic	Almost same to the primary focus
Peripancreatic lymph nodes	Red-bean size	
Urinary bladder	Many (about 33, from 3×3 mm to 10×10 mm) papillary nodules	
Left ureter	Irregularly papillomatous	papilloma
Colon	6 intestinal polyps (from red-bean to little fingertip size)	Adenoma
Thyroid	R. 50×25×30 mm nodule 4×4×4 mm L. 50×25×30 mm nodule 12×18×5 mm	Struma nodosa parenchymatosa papilliferum
Cortex of the right kidney	Round gray nodule (3×3mm)	Papillary adenoma from tubular epithelium
Right adrenal cortex	Soybean-sized nodule (3×3 mm)	Nodular hyperplasia of the cortex
Medulla of the right kidney	Grayish-white nodule of 4 mm in diameter	Fibroma
Prostate gland	Nodule of 14×14×4 mm	Fibromatous hyperplasia

gically removed under the pathological diagnosis of papilloma in the renal pelvis. Since 3 years ago, the patient complained of hematuria, pain by miction and dull pain in the lower parts of the abdomen.

One month before death, a circumscribed nodular shadow in the hilar portion of the right lung was found by X-ray examination.

140. ON THE MECHANISM OF THE TUMORIZATION WITH FEEDING TEST OF THE YOSHIDA TUMOR

KATSUO OGAWA, AKIRA TSUTSUMI and KATSUMI IWATA

(Dept. of Path., Med. School, Okayama Univ.)

Hamazaki *et al* previously reported that the diffuse tumorization, the swelling and blackening of Harderian gland were truly specific findings observed in the albino rats fed with Yoshida tumor. In the present experiment we have carried on investigations concerning the following problems.

1. Histological changes of the skull base at the early stage of tumorization: At an early stage the accompaniment of the infiltration of inflammatory cells with bleeding in perineural lymphatic spaces could be observed. Followed by the swelling and proliferation of Schwann's cells, gradually progressed the tumorization. This further extended to periphery and in the case of the first branch of trigeminal nerve the blackening of the Harderian gland seemed to occur. In the case of tumorization of the petrous part of the skull base showed the derangement of tissues due to mesenchymal cell proliferation in the loose connective around tympanum, and this ultimately presented a picture resembling reticulosarcoma. Even in the fairly advanced cases tumor cells could not be recognized in the tympanum.

2. Histo-chemical investigations of brown pigment in the Harderian gland: The pigment observable in the spaces of the gland was positive to melanine and Schmorl's lipofuchsine tests and did not respond to the pigments derived from blood. So far a considerable amount of porphyrine had been recognized to exist in this gland, but its relation with this pigment is now under our investigation.

3. Electron-microscope findings of the Harderian gland: A marked development of vesicular endoplasmic reticulum could be recognized. In addition, there could be detected many larger vacuoles containing lipoid bodies.

4. Electron-microscope findings up to the tumorization of lymph nodes: In the early stage of the tumor feeding nuclear inclusions were formed in the reticulum cells consisted of fine granules. In the cases of early tumorization such as those with

seven feedings a striking increase in endoplasmic reticulums in tumor cells could be observed. On the contrary endoplasmic reticulums had been scarcely proved in Yoshida tumor cells. (文部省科学研究費による)

141. TWO CASES OF CHOLANGIOCARCINOMA INDUCED BY THOROTRAST ADMINISTERED MORE THAN 20 YEARS AGO

YOHEI II, TSUGIO KATO, YOSHIYUKI MORIMURA,
KENJI NASU, TAKAYUKI YAMAUCHI and TORU MIYAJI
(1st Dept. of Path., Med. School, Osaka Univ; Institute for Cancer
Research, Med. School, Osaka Univ.)

Case I. A 48 year-old male school clerk was admitted to the hospital because of jaundice. He proved to be a fatal case of cholangiocarcinoma. At the age of 20, he was X-rayed for jaundice but he was not sure whether contrast medium had been administered then.

Case II. A 55 year-old housewife was hospitalized complaining jaundice. She was a fatal case of cholangiocarcinoma. At the age of 32 years, she was administered a contrast medium on the X-ray examinations for jaundice. She denied administration of any other contrast medium. On hospitalization plain X-films revealed in both cases reticular shadow in the hepatic and splenic region. Following administration of EDTA both cases demonstrated a radioactive substance with 10.6 hours half-life in the urine. It was assumed that the substance was Pb-212 decayed from Th-232. On autopsy, both cases revealed cholangiocarcinomas possibly of hepatic origin. Thorotrast particles were detected in RES cells and periportal connective tissues of the liver. They were found in RES cells, trabecules and follicles in the spleen. They were also found in the lymph nodes and bone marrow in Case I. Both cases showed diaphragmatic, bile duct, gall bladder and bilateral pulmonary metastases. The autoradiography revealed α track on the sliced liver. The above assumption was confirmed by the examinations using the γ -scintillation spectrometer. The particles observed in the liver and spleen proved to be Th-232 on the basis of the measurements of 2.61 Mev γ -ray from Tl-208.

142. STUDIES ON LYMPHATIC TUMOR OF RF MICE INDUCED BY IRRADIATION. (I)

SHOZO IRINO, TAKASHI KOTSUKA and MASAYOSHI KIBATA

(Dept. of Internal Medicine (Hiraki Clinic), Med. School, Okayama Univ.)

1) By the total body irradiation with the dose of 350 r by X-ray to 30-40 days old RF mice, leukemia has been successfully elicited in 73 per cent of the test animals, and the following findings was obtained.

2) In the mice with this experimental leukemia, there were those with a marked tumor-like swelling of the thymus (*thymic type*) and those with a slight enlargement of the thymus (*non-thymic type*), and these two groups showed distinct differences in the changes of peripheral blood picture and also in the manners of the infiltration of leukemic cells into the principal organs. Namely, in the thymic type Glisson's capsule itself showed almost no infiltration or only a slight one but a discrete infiltration mainly in the sinusoid adjacent to Glissons's capsule, whereas the non-thymic type revealed the infiltration in a nodular form around the blood vessels within Glisson's capsule.

3) By the clinical tissue culture devised in our department, the primary leukemic cell proliferation could be observed in the thymus in the case of the thymic type while the primary infiltration of these cells could be observed in the spleen or in the lymph nodes in the case of the non-thymic type.

4) The cytological observations had been conducted with fluorescence microscope and by the double vital staining with neutral red-Janus green.

5) And also in the transplantation experiment with the spleen and lymph nodes of the leukemic mice, it had been successful in all the cases tried.

143. ON THE RELATION BETWEEN THE ELECTRONIC STRUCTURE AND THE CARCINOGENIC AND CARCINOSTATIC ACTIVITIES OF URETHAN AND ITS RELATED COMPOUNDS

KEN'ICHI FUKUI, CHIKAYOSHI NAGATA, AKIRA IMAMURA,
and YUSAKU TAGASHIRA

(Faculty of Engineering, Kyoto Univ.; Institute for Virus Research, Kyoto Univ.)

The electronic structures of ethyl carbamate (urethan) and its related compounds are obtained by way of the frontier electron method which has been established by

the present authors.

The result of calculation is compared with experimental cancer-producing activities, and a distinct relation is found between the frontier electron distribution for nucleophilic attack at the position of the ester carbon atom and the carcinogenic activity of these compounds. This finding may add a support to our postulation that an interaction between the reactive site of carcinogenic compounds with nucleophilic center in the body might be involved in the genesis of tumors. In relation to this view, the carcinogenic activities of miscellaneous chemical carcinogens such as alkylating agents, acetylaminofluorenes, dulcin, thiourea, formaldehyde, etc. are discussed and a unified explanation on the carcinogenesis is given from the standpoint of nucleophilic reactivity of these compounds.

An interrelation between the carcinogenic or carcinostatic activity and cholinesterase inhibition of chemical compounds is searched for, and the identity of reaction nature of these physiological actions is pointed out. Accordingly, the carcinogenicity of various kinds of organic phosphorus insecticides which have both the anticholinesterase and anti-tumor activities is anticipated.

The reactive group or groups which are truly concerned with the production of tumors may probably be findable among the nucleophilic groups. Such groups may be specific with regard to the sterical situations.

The Present study is supported in part by the grant-in-aid of the Ministry of Japanese Government.

附 議

中原和郎：このような理論的な研究では、癌原性というような語を一樣に同価値のものと考えすることはどうかと思う。ウレタンのごときものはある人は癌原性と表現しているが、4-nitro quinoline N-oxide や methylcholanthrene のようなものが癌原性であるというのとは非常に違う（ウレタンで悪性腫瘍ができたためしはない）ことが注目されなければならない。

福井：芳香族炭化水素や4-ニトロキノリンN-オキシドなどと違ってウレタンのつくるものは一般に良性腫瘍であります。でき上った腫瘍としては悪性と良性のものは大きく異なるかも知れませんが、先生の御説にもありますように発癌機構の段階では同じである可能性が大きいと思われます。私達がみようとしているのはこの初期段階でありますので理論指数と比較してこの系列内で比較してみたわけであります。

寺山 宏：Carcinogen と求核反応性基との反応は一般にいて *in vitro* でも起るような unspecific な反応であるとするならばある種の carcinogen について非常に Organ や Species specificity が高いことはどう考えたらよいのか。

福井：臓器特異性、種属特異性の問題は非常に難しく、むしろこちらからおききたいところなのですが、一応私達は次のように考えているのですがいかがでしょうか。種属によりあるいは臓器によって代謝の様式が違うとともに、発癌物質の方も代謝に関係する代謝領域はいろいろ違っていると考えられ、しかもこのような領域は発癌領域とは異なっていると思われます。したがって発癌反応の様式はいろいろの発癌剤について同じく nucleophilic であっても、代謝領域の違いによって臓器、種属特異性が生ずる可能性があると考えます。

144. MOLECULAR ABNORMALITY OF A CERTAIN γ -GLYCOPROTEIN AND THEIR CHANGES DURING CANCER DEVELOPMENT

SEN'ICHIRO HAKOMORI, HIROAKI KAWAUCHI and TAIKO ISHIMODA

(Dept. of Biochem., Attached Institute for Cancer Research, Tohoku College of Pharmacy.)

An abnormal glycoprotein has been isolated from urine of patients with cancer as well as those of cancer bearing rats. It was found in the most carbohydrate rich but sialic acid poor fraction (Fr. 4) of benzoic acid adsorbable glycoprotein (I). "I" was dissolved in water at pH 8.4, precipitated at pH 4.5, the supernatant was electro dialyzed to remove precipitable glycoproteins and the supernatant lyophilized (Fr. 4). Fr. 4 was separated into several peaks on vertical zone electrophoresis. Peak 3 was the abnormal γ -glycoprotein (II), which was characterized by the following facts as compared with the normal γ -glycoprotein (III): 1) 3-4 times larger molecular weight, 2) 2-2.5 times greater number of none reducing terminals of oligosaccharide bunches, 3) decreased hexose/hexosamine ratio, 4) both "II" and "III" contained N-formyl-hexosamine besides N-acetyl-hexosamine and the ratio N-formyl/N-acetyl was greater in "III". The hexose/hexosamine ratio and the degree of branching of Fr. 4-glycoprotein isolated from rats urine has been followed up after being inoculated A.H. 130 and 7974. A marked changes has been recognized even at the earliest stage of cancer development and the ratio decreased, degree of branching increased more and more in compliance with the development of cancer.

Antibody formed on injection of "II" react with "II" but also with "III", however the former was inhibited by addition of N-formyl-glucosamine.

145. RELATION BETWEEN THE ENZYMATIC N-DEMETHYLATION AND THE CARCINOGENIC ACTIVITY OF VARIOUS AMINOAZO DYES

MITSUO MATSUMOTO and HIROSHI TERAYAMA

(Dept. of Biophysics & Biochem., Faculty of Science, Univ. Tokyo)

A method of spectrophotometric assay of formaldehyde in biological mixture has been reported by MacFadyen but the original method is unsatisfactory to the assay of the oxidative N-demethylation of aminoazo dyes with rat liver homogenate because of interfering products generated during the long heating. After a few modifications devised by us the formaldehyde could be easily estimated and the rates of the

N-demethylation among various aminoazo dyes were compared with the carcinogenic activity of them. The results obtained may be summarized as follows: 1) The aminoazo dyes hard to be N-demethylated have no carcinogenicity. 2) But the aminoazo dyes sensitive to the N-demethylase are not always carcinogenic (for example: 2-methyl-DAB). 3) The order in the rates of N-demethylation among various ring methyl substituted derivatives of DAB is 2'-methyl-DAB (*2-3) > 2-methyl-DAB (0) > DAB(6) > 3'-methyl-DAB (10-12) > 4'-methyl-DAB (<1) > 3-methyl-DAB (0). These dyes except 3-methyl-DAB showed almost similar behavior. 4) In the case of various N-substituted derivatives of MAB, the order was DAB (6) > N,N-methyl, n-propyl-AB (?) > N,N-methyl, ethyl-AB (6) > MAB (6) > N,N-methyl, isopropyl-AB (?) > N,N-methyl, benzyl-AB (0) > N,N-methyl, hydroxyethyl-AB (0). Both N,N-methyl, benzyl- and N,N-methyl, hydroxyethyl-AB were almost inert to the N-demethylase. 5) Upon comparing the rates of N-demethylation between DAB and MAB or their homologues, it was noticed that DAB (6) and 3'-methyl-DAB (10-12) produced about two-fold as much amount of formaldehyde than MAB (6) and 3'-methyl-MAB (10-12) under the same condition. 6) N,N-dimethylphenylenediamine (0) which seemed to be produced by azo-reduction of DAB did not produce formaldehyde under the same experimental condition. (*The figures in the brackets are the relative carcinogenic activities according to Miller and Miller).

A part of the expenses was covered by a grant from Ministry of Education of Japan.

146. SEPARATION AND PURIFICATION OF THE POLAR DYES

AKIRA HANAOKI, HIROSHI TERAYAMA*, SHOICHI KANDA*,
and MORIZO ISHIDATE**

(National Institute of Radiol. Science; Dept. of Biophysics and Biochem., Faculty of
Science*; Dept. of Pharmac. Science, Univ. of Tokyo**)

Polar dyes, which were prepared from the liver proteins of rat fed carcinogenic aminoazo dyes, were separated into four or five fractions by the method of ion exchange chromatography using Amberlite IRC-50. The polar dye of fraction 2, the main component, was assumed to be homogeneous and have the N-methylaminoazobenzene-like structure bound at 3-position of aminoazo dye moiety with the protein residue. However, its molar extinction coefficient at 520 m μ (in 2 N HCl-50% EtOH) determined by tracer method using 4-N-dimethylaminoazobenzene-¹⁴C (U) and by chemical analysis based on the determination of aniline derived from reductive cleavage of the polar dye at the azo-bond is 32000 and 45000 respectively. These

results may suggest that the polar dye (Fr. 2) is still heterogeneous and can be further fractionated by some methods. The authors tried to fractionate it by paper chromatography and paper electrophoresis and got at least two components; one is the polar dye previously suggested (1) and the other seems to be produced from (1) by oxidation such as demethylation and hydroxylation. The result obtained from the paper electrophoresis suggested the presence of the phenolic polar dye.

147. REACTIVITY OF 4-NITRO GROUPS IN THE REACTION BETWEEN 4-NITROQUINOLINE N-OXIDE AND ITS RELATED COMPOUNDS

TOSHIHIKO OKAMOTO and MICHIIYA ITO

(Faculty of Pharmac Sciences, Univ. of Tokyo)

4-Nitroquinoline N-oxide has been known to be a skin cancer producing agent, and Nakahara *et al.* have suggested that the cacinogenic action of this compound may be due to the nucleophilic substitution at position 4.¹⁾ In 4-nitroquinoline N-oxide and its related compounds, H. Endo reported that there was a certain relationship between the chemical structure and carcinogenic activity.²⁾ Furthermore, K. Fukui *et al.* proposed the similar relation between the electronic structure and carcinogenic activity.³⁾

	Fukui's theoretical index. Nakahara's Exp.			
	rate const $k \times 10^{-3}$ at 40°C.	approximate superdelocaliz- ability.	frontier electron density.	skin cancer producing activity.
4,6-dinitroquinoline N-oxide	3588	—	—	—
4,8-dinitroquinoline N-oxide	1512	—	—	—
4-nitro 7-chloroquino- line N-oxide	252	(3.361)*	(0.189)*	+
4-nitroquinoline N-oxide	70	3.113	0.186	+
4-nitroquinaldine N-oxide	17.0	1.913	0.178	+
4-nitroquinoline	8.3	1.579	0.176	—
4-nitropyridine N-oxide	6.4	1.092	0.149	—

*; 4-nitro 6-chloroquinoline N-oxide

1) Nakahara, W., and Fukuoka, F. Gann, 50: 1 (1959).

2) Endo, H. Gann, 49: 151 (1958).

3) Fukui, K., Imamura, A., and Nagata, C. Gann, 51: 119 (1960).

The following compounds react readily with SH-group of thioglycolic acid, and afford the corresponding 4-SR compounds.

With these nitro compounds, the present authors studied the nucleophilic reaction between 4-nitro group and thioglycolic SH- group, and determined the reaction rate constants, heats of activations and entropies of activations. According to this quantitative study, the activity values of 4-nitro groups show a good agreement with the theoretical results of Fukui's calculation. Furthermore, comparing these experimental results and Fukui's results with the carcinogenic activities, one would notice a definite relation between these chemical constants and the carcinogenic activities.

And, there seems to be a certain "upper threshold" as well as "the lower threshold" for the occurrence of carcinogenic activity.³⁾ But in this substitution reaction, the heat of activation and the entropy of activation are influenced by the composition of the reaction solution. With this result, if we assume 4-nitroquinoline N-oxide to react with the certain nucleophilic site of the biochemical system for the occurrence of the carcinogenic action, it seems to be difficult to estimate the carcinogenic activity by the chemical activity only of these compounds.

附 議

中原和郎：反応速度論的に癌原性の誘導体の最低のものと、非癌原性と認められる 4-Nitroquinoline との間の差はなほだいたい点が気になる。癌原性なしと見ている 4-Nitroquinoline でもあるいは多少の癌原性があるのかも知れない。発癌実験は粗雑な、あまり定量的でない実験なので、今後もっと精密にやってみると、反応速度論とさらによりよい一致を示すことがわかるのではないかと思っている。

伊藤道也：4-Nitroquinoline がその反応速度において 4-Nitroquinoline N-oxide の約 1/10 であるがこの反応速度の差と、発癌性の有無との間に問題があると考えられます。

もし N-oxide group のない 4-8-dinitroquinoline では 4-nitro group の反応性はかなり大に考えられますので、これらを今後合成し、その発癌性を検討する必要があると思われる。合成して検討する必要があると思われる物としては、4-8-dinitroquinoline, 4,6-dinitroquinoline, また pyridine 等で 4-nitro の activity が大になるような置換基をもつものが考えられます。

148. EXPERIMENTAL STUDIES ON DISPOSITION AND CARCINOMA (I) ALTERATION OF DISPOSITION AND BODY REACTION

KIKUJI TOKITA, SUKEAKI NISHIMUTA and KAZUKO SEKI

(Dept. of Pharmacology, Med. School, Toho Univ.)

The experiments were undertaken by altering the disposition of body to accerate to provoke or to inhibit either systemic reations including reflexes and shock or the changes after some heavy stress. The results of the experiments are as follows:

1) Bradycardia induced by digitalis or adrenaline did not appear after cutting vagal nerves. But when either continuous injection of acetylcholine or pretreatment with

nucleotide or chlorpromazine were made, the digitalis-induced bradycardia occurred intensively even after vagal nerves were cut. But the pretreatment of ATP did not involve the above facts.

2) In guinea-pig, the pretreatment of cortisone or egg-white accelerated the death by penicillin, but by the treatment of diphtherial toxoid, such death was completely prevented.

3) When croton oil was injected into muscle of uterus, 80-90% of the animal died. By the pretreatment with diphtherial toxoid, this death could be totally inhibited and the symptom was lightened.

4) When the teeth of rat were extracted, the animal died after 10 days. But when horse serum was injected 20 days before the extraction, they survived about 30 days and the symptom of tooth-extraction phenomena became slighter.

From the above facts, the systemic reaction which cannot appear in normal conditions, appears under some treatment. When some substances are applied to reflex arc, whose pathway is cut, the reaction which is observed in the intact arc, can be provoked, and after an animal is pretreated even with some substances which provoke no direct systemic reaction, they can accelerate or inhibit the systemic reaction. In conclusion, systemic reaction would be controlled by such alteration of disposition.

149. EXPERIMENTAL STUDIES ON DISPOSITION AND CARCINOMA (II) BODY REACTIONS IN YOSHIDA SARCOMA RATS

KIKUJI TOKITA, RYUTA ITO, HIRONORI KAWAMURA,
MITSUO YAMADA, YOSHIKO MIYAZAKI, HIROSHI HENOMATSU,
TOSHIO NAGAOKA, SHOSUKE UNEMOTO, CHIFUYU ISONO,
KUNIO TOSAKA, KEISHIO TAKASHIMA and KANJUN NAGATA
(Dept. of Pharmacology, Med. School, Toho Univ.)

Following studies were made for 8 days until the Wister rat died after Yoshida sarcoma had been implanted. Remarkable changes were observed in blood-picture. Decrease in number of erythrocytes appeared next day of the transplantation. The minimum level was reached after 3 days and this level continued till the animal died. Leucopenia was evident until 3 days and began to recover after the 5th day. Percentage of neutrophils increased evidently and continued till the animal died. Eosinophils decreased at the 3rd day, increased at the 5th day and again decreased before the death. 17 keto-steroid content in urine decreased next day of the transplantation, recovered at the 5th day. But K in urine disappeared and K in blood increased immediately before the death. From the above facts, it can be remarked that Yoshida sarcoma is not only an intensive stress but its products involves the

acute intoxication and the latter fact may suggest that the symptom may be prevented by the alteration of the disposition which reported in the first paper of these series of works.

150. EXPERIMENTAL STUDIES ON DISPOSITION AND CARCINOMA (III) PREVENTION OF CARCINOGENESIS BY THE ALTERATION OF DISPOSITION OF THE HOST

KIKUJI TOKITA, KUNIO TOSAKA and KANJUN NAGATA

(Dept. of Pharmacology, Med. School, Toho Univ.)

Studies were made to find the substances which can prevent carcinogenesis, when the substances were injected before the transplantation of Yoshida sarcoma. The substances examined were egg-white, diphtherial toxoid, γ -globulin, serpentina alkaloid, ATP, nucleotide, pantothenic acid, digitalis and sera of various animals, i.e. rabbit, guinea-pig, ox, horse, rat and human. Out of these substances, horse serum revealed the most intensive prevention not only against the carcinogenesis but also against tooth-extraction phenomena. The effective treatment was to inject 5 cc/body of horse serum every 5 days repeatedly for over 3 times 20 days before the transplantation. The mortality percentage of rats by Yoshida sarcoma was lowered to 10-50% (average 40%) after the pretreatment of horse serum. This obtained resistance in rat continued for 3 to 5 months and no evident changes were observed in organs. From the above facts, it can be remarked that some principles in that serum could raise the natural resistance not only against carcinoma but against tooth-extraction phenomena. This principle existed not only in the serum of horse but in those of other animals. The potency of the serum was stronger in the descending order: that of horse, ox, guinea-pig and O-type of human. And as is described in the next report, the repeated injection of that substance obtained from horse serum evidently raised that resistance and this substance did not provoke anaphylactic shock or Schultz-Dale's reaction not only in rat but in guinea-pig like desensibilization. This substance is named by Tokita, Natural Resistance Enhancing Substance (NRES).

附 議

滝沢延郎：馬血清の繰り返し注射によってラットの体重その他に変化はおきませんか。

武田：癌移植で治る治らぬは先ず動物の系液が問題である、演者の場合ラットの系液との関係は如何。

戸木田：滝沢教授に対して——ラットに馬血清を注射して何等の変化も見られません。体重の減少も見られませんし、モルモットのような anaphylaxy 現象も見られないし腸管の Schultz-Dale 反応も見られませんでした。

武田教授に対して——馬血清反覆注射による抵抗力増加は大沢ラットに対してはきわめて著明な効果 (70%) がありますがドンリウラットに対しては効果が少なく 30% の永久生存率を示しました。

151. THE EFFECT OF NICOTINAMIDE AND DIPHOSPHOPYRIDINE NUCLEOTIDE ON AZO DYE AND METHYLCHOLANTHRENE CARCINOGENESIS

MUTSUSHI MATSUYANA and TAKEO NAGAYO

(Dept. of Path., School of Med., Nagoya City Univ.)

Using rats, the authors studied the effects of nicotinamide upon the incidence and growth of experimental tumours induced by p-dimethylaminoazobenzene (DAB) and 20-methylcholanthrene (MC). The effect of diphosphopyridine nucleotide (DPN) on the incidence of MC sarcoma was also examined additionally.

Experiment using DAB as carcinogen....A total of 55 rats of Sprague-Dawley strain were used. Group 1: Basal diet+Nicotinamide inj. (15 rats). Group 2: DAB diet +Water inj. (15 rats). Group 3: DAB diet+Nicotinamide inj. (25 rats). In groups 1 and 3 animals were injected subcutaneously with 0.25 ml of 1% nicotinamide, while in group 2 animals were injected equal volume of distilled water. Injections of three groups were started 1 week before commencement of azo dye feeding, and continued 3 times weekly. All animals which were fed with dye diet and survived the experimental period over 100 days developed hepatoma. The average liver index of the animals of azo dye feeding groups were; 24.0 in group 2, 22.7 in group 3, respectively.

Experiment using MC as carcinogen....A total of 36 Wister strain rats weighing about 125 g were used. Group 1: MC inj.+Water inj. (12 rats). Group 2: MC inj.+Nicotinamide inj. (12 rats). Group 3: MC inj.+DPN inj. (12 rats). Injections of nicotinamide and DPN (0.1%) were performed in the same manner as in the first experiment. One week after the first injection, all rats were injected subcutaneously in intrascapular region with 3 mg of MC in 0.4 ml of olive oil. The time of "100 per cent tumour incidence" in each group was; 120th day in group 1, 168th day in group 2, and more than 168th day in group 3. Furthermore, the tumour-incidence rates in groups 2 and 3 were still 30 and 36 per cent respectively, even when that in group 1 reached 100 per cent. Average value of tumour indices in each group was as follows, 28.1 in group 1, 17.8 in group 2, and also 15.1 in group 3. From these results, it may be said that the incidence and growth of MC sarcoma was delayed by administration of nicotinamide and DPN. In the case of DAB liver cancer, however, the inhibitory effect was very slight.

This work was supported in part by reserch grants from the Ministry of Education and from the Japan Cancer Society.

**152. DEVELOPMENT OF LIVER CANCERS IN THE RAT BY
4-DIMETHYLAMINOSTILBENE (DAS) FEEDING FOLLOWING
INITIAL 4-DIMETHYLAMINOAZOBENZENE (DAB) FEED-
ING. (I) EXAMINATION OF THE RAT FED INITIALLY
WITH DAB FOR MORE THAN ONE MONTH**

SHIGEYOSHI ODASHIMA (The Med. Institute of Sasaki Foundation, Tokyo)

4-Dimethylaminostilbene (DAS) was reported by Haddow as a strong carcinogenic substance for developing ear duct cancer in the rat. The present report deals with the effects of DAS-feeding on the development of liver cancers in the rat following initial 4-dimethylaminoazobenzene (DAB) feeding.

Two hundred and fifty-nine male Wistar rats were employed. They were divided into 8 groups and fed with semisynthetic diet containing DAB (0.06%) or DAS (0.005%) for 5 months as the way shown in the table, respectively. After the stop of the feedings, they were fed with the basic diet and examined for the development of cancers throughout 360 experimental days. The results obtained were as follows (See also the table)

Groups	Mos. of feeding DAB—DAS		Number of animals		Liver cancers	
			Employed	Valids	No. of rats	%
I	5	0	23	22	17	77
II	a	4	23	23	18	78
	b	4	9	8	5	63
III	a	3	22	19	19	100
	b	3	5	4	2	50
IV	2	3	21	19	18	95
V	1	4	28	28	19	68
VI	0	5	128	92	7	8

1. In DAS-group (Group VI), 82 tumor rats containing 75 cases of ear duct cancers, 7 of liver cancers and 6 of mammary cancers were took out of 92.

2. In DAB-DAS-group, i.e., Groups II-a, III-a, IV, and V, the incidence of liver cancers took 78%, 100%, 95%, and 68%, respectively. The rate is similar or much higher than that of DAB-group (Group I), but distinctly higher than than of DAS-group (Group VI).

3. The average latent time of tumor incidence was shorter in DAB-group than those of DAB-DAS-group and it was about 8 months.

4. The average number of the tumor nodules induced in the liver was 3.2 in DAB-group. The number was more than those of DAB-DAS-groups.

5. The formation rate of lung metastasis observed in the tumor rats was 71% in the DAB-group. The rate was much higher than those of DAB-DAS-groups.

**153. ON THE HISTOGENESIS OF THE EAR DUCT AND LIVER
CANCERS IN THE RAT FED WITH 4-DIMETHYL-
AMINOSTILBENE (DAS)**

SHIGEYOSHI ODASHIMA (Med. Institute of Sasaki Foundation)

In 1947, Haddow reported 4-dimethylaminostilbene (DAS) as a strong carcinogenic substance for developing external ear duct cancers in the rat. The present paper deals with the histogenesis of the tumors and the liver cancers in the rat fed with the substance.

Two hundreds male Wistar rats were employed in this experiment. They were fed with semisynthetic diet containing 0.005% of DAS for 5 months and after the stop of the feeding, they were fed with the basic diet.

For studying the histopathological changes occurred in the liver, spleen, lungs, ear ducts and lymph nodes, five rats were sacrificed respectively, every one or two weeks from the beginning of the feeding to 200th experimental day. The remaining rats were examined for the development of cancers throughout the 360 experimental days. The results obtained were as follows:

1. Proliferation of squamous epithelium of external ear ducts was observed after 2 or 3 weeks of the feeding. After 2 months, papillomas appeared. Squamous cell carcinomas began to appear at 186 experimental days and 75 cases of the cancers were took out of 92 valid cases throughout the experiment.

2. Marked reticulosis was observed in the spleen and lymph nodes in almost all the animals.

3. Distinct proliferation of bronchial epithelium and 2 cases of adenomatous growth of alveolar epithelia were detected in the lungs.

4. Inter- and intra-acinar proliferation of bile canaliculi, cholangiofibrosis, cysts, and cirrhosis were observed as was the cases fed with 4-dimethylaminoazobenzene (DAB). Special findings of the liver differing from the DAB-rats are that small hemorrhagic fields and necrosis of liver cells were found sporadically, and that marked proliferation of liver cells was detected mainly in the peripheric region of the acini. The nodules presenting the histological picture of what is called adenomatous hyperplasia of liver cells began to appear at 5 months. Seven cases of liver cancers were detected out of 92 rats.

5. Six cases of mammary adenocarcinoma were also found at the end of the experiment.

154. ON ENZYMATIC AND CYTOLOGICAL CHANGES DURING THE CARCINOGENESIS OF MICE AND RATS (I)

KENWO ASANO (Dept. of Internal Med., Med. School, Okayama Univ.)

The lactic dehydrogenase (LDH) activity has been observed to be increased in the sera of experimental animals and in humans with malignant neoplasm.

This communication reports the results of the observations of LDH activity in the sera and in the tissue culture media.

Material and method: Strain C₃H mice were given 0.1 cc of 40 percent solution of carbon tetrachloride in olive oil or 0.1 cc of olive oil repeatedly 3 times weekly for 10 to 50 doses by stomach tube. Rats were fed on diet containing 0.06 percent DAB. Liver and cancer tissues were cultured by roller tube, and the determination of LDH activity was done spectrophotometrically.

Results: The serum LDH activity of C₃H mice treated with olive oil and carbon tetrachloride did not elevate. Therefore, no apparent relationship appeared to exist between the serum LDH activity and the dose of carbon tetrachloride or olive oil, although the elevation of activity appeared in some mice given 50 doses of carbon tetrachloride. The experimentally induced hepatoma bearing C₃H mice showed significant elevation of the serum LDH activity.

The livers of normal C₃H mice showed slight rise of LDH activity in cultured media. The rise of activity in media bathing livers treated with olive oil was not different from the normal mice. The LDH activity in media bathing hepatomas rised significantly and rapidly, and the activity of media bathing livers treated with carbon tetrachloride of 50 doses rised moderately on 6th day.

The serum LDH activity of DAB fed rats was examined every one month. The activity gradually elevated until the occurrence of liver cancer, and tumor bearing rats showed significant elevation of the serum LDH activity.

An apparent relationship appeared to exist between the duration of DAB feeding and the activity in cultured medium bathing liver from DAB fed rat. The activity of LDH in medium bathing liver cancer tissue showed significant rise rapidly.

附 議

勝田：組織片培養では各培養管あたりの細胞数を一定にできないので、それらの活性を比較しても、今日の研究レベルから見ると何もいえない。初めにトリプシナイズか何かして、細胞浮游液として接種し、一定細胞数当りの活性値で比較することをおすすめる。

浅野：① 培養液は作製時に活性値にかなり差があるため、毎回ブランクを作製して、培養時の活性値に対する比を求めた。

② 私達は臨床的に困難な肝臓癌の早期診断を意図し、そのための培養に簡便な初代培養を使用している。

**155. HISTOCHEMICAL STUDIES DURING CARCINOGENESIS IN
MOUSE SKIN INDUCED BY 20-METHYLCHOLANTHRENE
(I) THE LOCALIZATION OF ALKALINE PHOSPHATASE,
ACID PHOSPHATASE, ESTERASE AND β -GLYCOSIDASE
DURING CARCINOGENESIS**

KENSAKU KAWAKATSU, MASAHIRO MORI, YOSHIHIRO OKAMOTO
and RYUICHI OKA

(Dept. of Oral Surgery, Dental School, Osaka Univ.)

The histochemical demonstration of alkaline phosphatase, acid phosphatase, esterase and β -glycosidase in the process of experimental carcinogenesis in mouse skin has been carried out. A total of 100 mice received applications of 0.5% 20-methylcholanthrene in acetone delivered to their backs by a brush twice a week up to 21 weeks. Of these mice, 6 were sacrificed every other week, and carcinogenic process in various stages was observed. Fresh frozen sections of the skin with a thickness of 10 to 20 μ were cut in the cryostat at -20°C . At the same time, hematoxylin-eosin stained preparations were made from frozen sections to serve as histopathological findings.

No alkaline phosphatase activity was observed in cancerous tissues of various stages and in normal skin, while as epidermal hyperplasia progressed and the connective tissue proliferated, the enzyme was abundant in the connective tissue adjacent to the infiltrative portion of cancer cords.

Acid phosphatase of experimental cancer was present in the granular layer, moreover, in the hornified layer, the epithelial pearls and the hair follicles. In the normal skin of mice, this enzymatic activity was observed in the superficial layer and the hair follicles.

Esterase of the cancerous tissue was rich in the epidermis, and in normal tissues its activity was found also in the epidermis.

The localization of β -glucuronidase, β -glucosidase and β -galactosidase was similar each other. These enzymes were contained in the granular layer, the epithelial pearls and the hair follicles of skin carcinomas. The most striking reaction of β -glycosidase of normal skin was found in the hair follicles and the fat tissue, and slight reaction was observed in the epidermis of the skin.

The distribution of acid phosphatase, esterase and β -glycosidase in normal tissue and skin carcinoma was nearly the same.

**156. HISTOCHEMICAL STUDIES DURING CARCINOGENESIS IN
MOUSE SKIN INDUCED BY 20-METHYLCHOLANTHRENE
(II) HISTOCHEMICAL DEMONSTRATION OF PROTEIN-
BOUND SULFHYDRYL AND DISULFIDE GROUPS AND
PAS REACTIONS DURING CARCINOGENESIS**

**KENSAKU KAWAKATSU, MASAHIKO MORI, YOSHIHIRO OKAMOTO
and RYUICHI OKA**

(Dept of Oral Surgery, Dental School, Osaka Univ.)

One hundred mice were painted with 20-methylcholanthrene 0.5% in acetone on the skin of the back after cutting of the hair twice a week. The experimental period in various stages was about 21 weeks. 6 mice were killed every other week, and fresh frozen sections were cut at 10 to 20 μ in the cryostat at -20°C . These sections were fixed with 80% alcohol for 10 minutes, and used for the demonstration of protein-bound SH and SS groups.

Hematoxylin-eosin was used for staining preparations. Several sections were also fixed with 80% alcohol, embedded in paraffin, and the PAS stain was employed for polysaccharides. The neotetrazolium method of Gomori for protein-bound SH and SS groups was used. Fresh frozen sections were dried at room temperature, then fixed in trichloroacetic acid alcohol for 20 minutes, rinsed briefly in distilled water, and incubated in the following solution for 8 to 12 hours at 37°C : 1) 10% KCN (10ml), 2) 20 ml 0.5 M Borate buffer pH 8.5 (20 ml). A drop of phenolphthalein solutions was added and titrated with 0.1 N acetic acid until completely discolored, and then added neotetrazolium chloride (25 mg). After incubation, these sections were washed in 1% acetic acid, and mounted with glycerol. Thioglycolic acid was used as the reducing reagent of the SS bridge. Tissue sections were immersed in the following mixture for 2 hours at 37°C : 0.4 M 10 ml thioglycolic acid was titrated to pH 8 with 0.1 N NaOH about 10 ml.

In normal skin, protein-bound SH and SS groups were contained in the epidermis and the hair follicles. In skin carcinomas, a high concentration of SH and SS groups was mainly present in the granular layer, the keratotic layer and the epithelial pearls.

PAS positivity was not found in skin carcinoma.

**157. HISTOCHEMICAL STUDIES DURING CARCINOGENESIS IN
MOUSE SKIN INDUCED BY 20-METHYLCHOLANTHRENE**

**(III) HISTOCHEMICAL DEMONSTRATION OF AMINO-
PEPTIDASE DURING CARCINOGENESIS**

KENSAKU KAWAKATSU, MASAHIKO MORI, YOSHIHIRO OKAMOTO
and RYUICHI OKA

(Dept. of Oral Surgery, Dental School, Osaka Univ.)

A total of 100 mice was used, and the skin of the back was painted with the solution of 0.5% 20-methylcholanthrene in acetone. All mice were painted twice a week for 21 weeks, and 6 were sacrificed every other week. Aminopeptidase activity in various stages was observed with the method of Nachlas, Crawford and Seligman. The skin was removed and fresh frozen sections approximately 10 to 20 μ thick were cut in the cryostat at -20°C . The sections were then fixed with 10% neutral formalin for 10 minutes after drying at room temperature, rinsed in water, and incubated in the following solution for 30 to 60 minutes at 37°C : 1) 8 mg/ml L-leucyl- β -naphthylamine hydrochloride (1 ml), 2) 0.1 M acetate buffer pH 6.5 (10 ml), 3) 0.85% NaCl solution (8 ml), 4) 2×10^{-2} M KCN (1 ml), and added diazo blue B (10 mg). After incubation, sections were rinsed in water, immersed in 0.1 M copper sulfate for 10 minutes, rinsed briefly in water again, dehydrated in graded alcohol, and mounted in balsam.

The portion of the enzyme activity was stained blueish purple. Aminopeptidase was contained in the basal cells and the hair follicles in normal skin. Aminopeptidase activity was not seen in skin carcinomas, but an intense reaction was observed in the connective tissue adjacent to the invasive proliferating epidermis and also in infiltrated mast cells.

**158. CHANGES IN ULTRASTRUCTURES OF RAT LIVER CELLS
DURING DAB CARCINOGENESIS**

**(II) SIGNIFICANCE OF CHARACTERISTIC CHANGES
IN ENDOPLASMIC RETICULUM**

TAMENORI ONOE, ISAO SUZUKI, GOHEI TAKAHASHI,
YAHEI KOSEKI and MASAKI NORO

(Dept. of Path. and Cancer Research Institute, Sapporo Med. College)

In the previous report, development of clumps of finely vesicular endoplasmic reticulum was noted as one of the characteristic changes of rat liver during DAB carcinogenesis.

Further studies revealed the occurrence of those structures in the very early stage during both DAB and 3'-Me-DAB feeding. For the latter, dye dissolved in olive oil was given by stomach tube in order to expect quantitative administration of the carcinogen. Electron microphotographs were prepared by conventional procedures.

Using 3'-Me-DAB, distinct clumps of finely vesicular agranular endoplasmic reticulum began to appear after 2 days of feeding. The clumps increased the area in the cytoplasm with the progress of feeding. In the early stage, irregularity of mitochondrial in shape and size and decrease in Palade granules attached to the endoplasmic reticulum were also noticeable. Biochemical study in the same stage revealed a decrease in RNA content without a decrease in total nitrogen and an increase in glycolytic activity.

Strikingly rapid decrease of clumps of vesicular endoplasmic reticulum was observed by leaving the animals from dye administration for 7 days after 7 days of dye feeding. At the same time the recovery of RNA content was observed biochemically. Meanwhile, glycolytic activity on the same time showed only a slight decrease, much higher than the activity of control rat liver, thus, the recovery of increased glycolysis to normal level delayed for longer than that of RNA.

From above findings it may be summarized that an increase in glycolysis and other evidences observed in endoplasmic reticulum have different significances, although all of those changes are considered to be specific in morphology and metabolism of rat liver cells caused directly with 3'-Me-DAB.

附 議

寺山 宏：3'-Me-DAB 投与の際に Palade Particle (RNA-Protein) が減少し、小胞体の変形がみとめられるということに関連して、われわれは RNA-Protein Particle と Azodye が強く結合していることを見出している (Nature 1960, 生化学会総会 1960)。このように Azodye に結合した RNA-Protein の運命に関して興味深い知見が与えられたものと考えられる。

小関：分割した細胞画分については結合色素量を測定しておりませんが、今後その点も検討したいと存じます。

村松正実：今の電顕像でちょっと見過したのですが mitochondria はどうだったのでしょうか。確か Porter らは初期にはあまり変化がないといっていたようですが……。

私達の所でも 3'-Me-DAB 飼育ラットについて代謝の変化を研究しておりますが、確かに非常に早期から glycogen の減少が認められます。しかしビルビン酸の TCA サイクルを通しての、代謝、特にその周辺アミノ酸への利用は2週間程度の早期にはあまり障害されないようです。そして2-3カ月の晩期に強く障害されてくる。もちろん肝癌では、利用は非常にわるい。これは Porter らの mitochondria の観察と一致するように思われたのですが、いかがでしょうか。

小関：ミトコンドリアにおいても DAB および、3'-Me-DAB 飼育の極く初期に著明な変形を認めており、呼吸代謝の変動もある種のものについては知見を得ているが、詳しい検討は今後に行う予定であります。

小関：小管状無顆粒子胞体の集簇の出現については私どもも DAB 以外の他の物質による肝病変または、十数時間の絶食時の肝においてもみとめているが、その程度、量、時間的な推移から DAB 飼育に特別なものの一つと考えたい。

**159. ON THE RELATIONSHIP BETWEEN CONGO RED INDEX AND
QUANTITATIVE HISTOLOGY IN THE RAT LIVER DURING
CARCINOGENESIS BY AZO DYES**

NOBUYUKI ITO, YOSHIAKI FUKUOKA, MASAO MARUGAMI,
HIROSHI NAKAMURA and HAJIME KITAMURA
(Dept. of Path., Nara Med. College)

The studies in this report were concerned with the relationship between congo red index (C.I.) and quantitative histology in the rat liver during carcinogenesis by azo dyes.

In the first series of experiments, a total of 210 adult male albino rats (Wistar strain, 180-240g) were used. The functional test of reticulo-endothelial system (RES) was based on Yasuoka's modification method of Adler-Reimann's. Then, rats whose C.I. showed under 20 or over 40 were excluded from the total number of rats. In the second series of experiments, 95 rats that had been selected by the above method were used. They were divided into 4 groups including that of the control. Each group was ingested with following; group 1, 0.06 per cent 3'-methyl-4-dimethyl-aminoazobenzene diet: group 2, 0.1 per cent o-aminoazotoluene diet: group 3, 0.1 per cent methyl-orange diet: group 4, basal diet. After certain weeks (2, 4, 6, 8, and 10), they were decapitated. The histological examinations of the liver were based on routine work and on Daust's method.

In this observations, the azo dyes which had high potency as liver carcinogen gave severe disturbances on functional activity of RES. It was concluded that the relationship between congo red index and numerical cell population of various cell types in the rat liver was not remarkable. Histological findings were very similar to those reported by various observers. However, the results indicated the existence of some relationship between the C.I. and the average percentages of PAS-positive reticulo-endothelial cells in the liver. From the above results, the destruction of azo dye in the hepatic tissue was weaker in the initial stage than that of the azo dye with high carcinogenic activity compared with that in low or non carcinogenic substances. Therefore, the concentration of azo dyes in the hepatic tissue was remarkable in the high potency carcinogen treated rats.

160. STUDIES ON THE PYRUVATE METABOLISM DURING EXPERIMENTAL HEPATOCARCINOGENESIS.

(I) IN VIVO METABOLISM OF PYRUVATE-2-¹⁴C

MASAMI MURAMATSU, TOSHIAKI OSUGA, SACHIO TAKASU,
YOSHITAKA ARAKI and SADATAKA TASAKA

(Tasaka's Clinic of Internal Medicine, School of Med., Tokyo Univ.)

Busch *et al.* studied *in vivo* metabolism of pyruvate-2-¹⁴C using transplantable tumors, and reported the strongly diminished incorporation of the ¹⁴C isotope into the TCA cycle-related amino acids in the tumor tissues as compared with many control tissues. They interpreted this fact by the lowered activation and condensation of pyruvate partly due to the depletion of oxaloacetate and by the hindered amination reaction from those compounds of TCA cycle. The authors wanted to confirm this fact in the 3'-Me-DAB induced hepatoma, and tried to follow up the change in the metabolism during hepatocarcinogenesis. Male albino rats of Wistar strain weighed 150-200 mg were fed rice contained 0.06% 3'-Me-DAB. At 3 weeks—the period which the uptake of this dye into the liver was maximum, 2-3 months—the stage of liver cirrhosis or precancerous state, and after the hepatoma emerged, the animals were injected 5 μ C of sodium pyruvate-2-¹⁴C intravenously. 3 and 8 minutes after the injection, the animals were decapitated and the liver excised and prepared for chromatography. The preparation of the tissues and the technic of anion exchange chromatography were approximately the same as those of Busch *et al.* We isolated alanine, glutamic acid, aspartic acid, lactic acid, succinic acid, and malic acid by this chromatographic technic, and measured the incorporation of ¹⁴C isotope in each acid.

The results were as follows: 1) in hepatoma, the uptake of pyruvate into the tissue and the metabolism through TCA cycle into amino acids were strongly suppressed as compared with normal liver.

2) In the carcinogenesis by 3'-Me-DAB, these changes arose in the late stage—cirrhotic or precancerous stage, but not in the early stage. For example, the incorporations into aspartic acid were 1690, 1430, 155, 5 c.p.m./1 gm. tissue in normal, early stage, late stage and hepatoma, respectively.

Although gradual alteration of metabolism was observed during carcinogenesis, there seemed to exist a gap between precancerous liver and hepatoma.

161. PAPER ELECTROPHORETIC STUDIES ON ASPARAGINASE IN THE LIVER OF RATS FED 4-DIMETHYLAMINO- AZOBENZENE (DAB)

TSUNEO SATO, YUTAKA TAMURA and SANJI KISHI

(Dept. of Biochem., Showa Med. College)

Semiquantitative investigation of asparaginase in the liver of rats fed DAB has been proceeded by introducing paper electrophoresis, in order to detect the possible visualization and certification of the two sorts of asparaginase, namely, Greenstein's asparaginase I and II.

Supernatant fluids of hepatic tissue homogenates of normal and DAB rats were introduced onto the paper of the apparatus. At the time the electrophoresis has been finished, the paper was cut into segments and each of them transferred to a Conway unit for the determination of the enzyme activity.

The activity pattern was then represented graphically as follows: in the ordinate was taken liberated ammonia-N γ and in the abscissa the actual migration distance of the enzyme in cm.

The pattern of the normal liver showed two prominent peaks, one at the part of faster migrating (peak I) and the other at that of an intermediate rate (peak II), when the used digestion medium was containing pyruvate and phosphate simultaneously. If there was contained no phosphate in the medium the peak I diminished markedly and in the case of pyruvate the peak II.

The authors postulated hereupon as follows: the peak I and II are perceived to correspond to Greenstein's asparaginase I and II respectively. This argument was strengthened by the result of heat-inactivation test, that the peak I was relatively heat-labile in contrast to peak II.

The pattern of cirrhotic liver was quite similar to that of normal liver except the peak I was slightly lowered. In the pattern of the liver of rats under continuous DAB feeding (in the 4th week) the peak II remained prominent while the peak I almost disappeared. In the pattern of hepatoma, resembling those under DAB feeding, peak II was yet observed even low but the sign of the peak I no more.

This investigation was supported in part by a grant from the Japanese Ministry of Education.

附 議

寺山 宏: 肝癌周辺の非肝部について Asparaginase Pattern を検索されませんでしたか。

DAB feeding で One month というのは丁度非常な critical な時期に相当しそれからあとで肝細胞は大別2つの方向に変わり、大部分は適応的にもとの正常時に近い状態に変化し、ごく僅かの cell が癌的变化をされると考えられるからです。

佐藤: 肝癌周辺の部のアスパラギナーゼ Pattern については測定を行っておりません。

162. METABOLIC PATHOLOGY OF RAT LIVER DURING DAB CARCINOGENESIS (II)

YAHEI KOSEKI, YOICHI JINNOHARA, TERUYUKI HIROTA,
AIKO INOUE and HIROSHI HOMMA

(Dept. of Path. and Cancer Research Institute, Sapporo Med. College)

In the previous report metabolic characteristics of rat liver during the early stage of DAB carcinogenesis were described (Gann, Vol. 50, Suppl. 1959). Among them very early occurrence of an increase in glycolytic activities was particularly noted.

The present study concerns further studies on metabolic changes of rat liver during the early stage of 3'-Me-DAB carcinogenesis.

Animals fed with conventional basal diet were divided into two groups. Each animal of I and II groups was administered 9.6 and 4.8 mg of daily dosis 3'-Me-DAB, respectively, divided twice a day. Stomach tubing was done in order to expect quantitative administration of the carcinogen.

In I group, severe degenerative changes were observed in the liver, although 9.6 mg of the dye are about equivalent dosis with that contained in the usual daily diet for DAB carcinogenesis. This means the way of entry of the carcinogen influences greatly on the course results of carcinogenesis.

Estimation of contents of azo-dye, total nitrogen, nucleic acids and lactate, and of oxidative and glycolytic activities was made at various intervals of the feeding.

Among the results, amount of dye contained in the liver reached 17 micrograms/gram tissue in I group on the 8th day of feeding, followed with rapid decrease by leaving animals from dye administration. In II group, dye content, which was 3.6 micrograms/gram tissue after feeding for 7 days, decreased to less than 1 microgram within next 7 days without dye administration. RNA content of the liver was reversely proportional with amount of the dye in the tissue in the early stage: instant decrease by dye administration, more marked in I group than the other, and rapid recovery after finishing dye administration. By electron-microscopic study, clumps of finely vesicular smooth-surfaced endoplasmic reticulum, which began to appear after dye-administration for only a few days and became marked for 7 days, mostly disappeared without dye-administration for 7 days.

Anaerobic glycolysis showed a marked increase with or without the addition of DPN in both groups after dye-administration for 7 days. By leaving the animals from dye-feeding for 7 days, a slight decrease, much higher than in control, was observed.

Although these findings were thought to be specific direct effects on the metabolism of rat liver for 3'-me-DAB feeding, some discrepancies noted above suggested different significances in DAB carcinogenesis.

163. SURVEY ON THE SPONTANEOUS MOUSE TUMOR OF INBREED FEMALE MICE OF DD-S HOMOGENOUS STRAIN

TERUO MINESHITA, TORU IWAKI, JINSAKU MAEDA,

KENJI YAMAGUCHI and KIYOKAZU NAGAI*

(Shionogi Research Laboratory,* 2nd. Dept. of Pathol., Osaka Univ.)

A survey on the spontaneous tumor of dd-s homogenous strain mice in our farm was made on approximately 5500 breeders over a period of one and a half years.

1. Occurrence percentage of tumor appearance among the total inbred female mice was 0.073%. The occurrence differs according to the number of parturitions, the percentage increasing after each parturition.

2. Percentage of appearance of the tumor according to location was highest in the inguinal area with 36%, followed by the axillar area 24%, abdominal 15%, perineal area 13% and cervical area 11% of all the cases.

3. Histological investigation of 61 of the mice tumors showed 49 to be adenocarcinoma probably originating from the mammary gland. These could be further divided into the differentiated type with 38 cases and undifferentiated type with 11 cases. Two were adeno-acanthoma, one a papilloma, one a cavernous angioma, and the other eight were inflammatory change of the lymphnode.

4. Transplantability of all the spontaneous tumors was examined using intact mice of the same strain. One tumor which at present has been passed up to 12 generations originally was the undifferentiated type of adenocarcinoma, but now it has changed into a rather anaplastic type with many mitotic cells.

5. Response to anticancer drugs greatly differs even in tumors of close similarity in their histological pictures.

附 議

螺良義彦：今日各地でマウスが維持されているので、それらの発癌率がちがってきているかどうかを知りたいと思っています。マウス乳癌の発生は月齢と関係がありますので、発癌率はそれぞれのマウスをどの月齢まで観察されているかによって異ってきます。それ故各地の dd マウスとの比較を容易にするために、観察期間がどの位でこの発癌率があったかをお尋ねします。

螺良義彦：今一つは経産マウスと処女マウスについて観察期間が違っていないかということです。同じ dd マウスで阪大徹研の川俣教授の維持している ddO マウスは処女マウスも経産同様の高発生率であります。dds が果してほんとうに処女発癌率が低いものであれば、ddO と異った性質になっているわけでありまして、果してほんとうに相異しているのかをお尋ねします。

演者：1. 動物の年令との関係は、年令という表現では表わしていないが経産回数で表わしておる。大体 2 カ月に 1 回分娩しているのでおよそその処は年令に換算することができる。後ほど調べてみることにする。

**164. FRAMKALLNING AV UNDERHUDSSARKOM HOS RÅTTOR MED
UPPREPADE INSPRUTNINGAR AV VATTENLÖSNING AV
STREPTOMYCIN- OCH DIHYDROSTREPTOMYCIN
SULFAT KOMPLEX**

KOTARO WARABIOKA (Cancer Institutet)

I allmänhet, tar det lång tid att framkalla cancer i djurförsök. För att minska förlusten av försöksdjur under försöken, som förorsakas genom sådana tillstötande sjukdomar som infektion m.m., ingivas ofta antibiotikum, t. ex. penicillin eller streptomycin.

Författaren utförde experiment med upprepade insprutningar av vattenlösning av streptomycin- och dihydrostreptomycin-sulfat komplex i råttor för att undersöka den cancerframkallande verksamheten av detta antibiotikum. 5% vattenlösning av streptomycin komplex (0.5 g streptomycin sulfat samt 0.5 g dihydrostreptomycin sulfat upplöses i 20 ml. destillerat vatten) insprutades i underhudsvävnaden på det högra landområdet hos 10 råttor 6 gånger i veckan. Koncentration och volym av insprutat lösning är nedanstående:

från början till ett halvt år, 50 mg/1.0 ml,
från ett halvt år till ett år, 75 mg/1.5 ml,
från ett år till ett och halvt år, 100 mg/2.0 ml,
efter ett och halvt år, 125 mg/2.5 ml.

Fibrosarkom framkallades hos 2 av 7 råttor, som överlevde mer än ett år. En råtta av hankön dog under 17: de månaden med ett sarkom av ett litet hönsäggs storlek på insprutningsstället och en råtta av honkön dog under 20: de månaden med ett sarkom av ett vaktelsäggs storlek. I ett fall en råtta av honkön, som dog under 20: de månaden med ett granulom av tumfingerspets storlek på insprutningsstället, observerades en sköldkörtel cancer av ett risgryns storlek på vänstra loben samt adenomatös hypertrofi av vänstra parasköldkörteln. Alla 4 de andra råttorna dog med granulom på insprutningsstället.

Nefrotiska förändringar av olika grader observerades hos alla råttorna.

I jämförelse med mina föregående redogörelser över framkallning av underhudssarkom hos råttor med upprepade insprutningar av destillerat vatten eller olivolja, finns det inga särskilda skillnader i fråga om vävnads reaktion, utvecklings-perioden samt sarkomframkallnings förhållanden i detta försök.

Det torde kunna nämnas att sarkomframkallning i detta försök med streptomycin komplex är ett slags fenomen av ospecifik cancerframkallning; dvs., det beror inte på streptomycinets särskilda kemiska reaktion utan på ospecifik störning av lokala underhudsvävnaden genom kroniska retningar.

165. EXPERIMENTAL CARCINOMA OF THE UTERINE CERVIX IN C₃H MICE BY 3,4-BENZPYRENE

KAZUSHIGE HIGUCHI, TSUTOMU HOSOKAWA, MASA HARU IWATA,
ICHIO SHINOZAKI, TADAMASA ARIHIRO and MASAKI AOYAGI
(Dept. of Obst. and Gyne., School of Med., Tokyo Jikei Univ.)

Induced carcinoma of the uterine cervix in mice has been produced by numerous investigators in foreign countries, but there are only a few in Japan.

The authors studied on the experimental carcinoma of the uterine cervix in C₃H mouse according to Scarpelli and Haam's reports.

Ninety-seven C₃H female mice, 3-7 months of age, were painted intravaginally twice weekly with 1% solution of 3,4-benzpyrene in acetone through infant-sized otic speculum. Twenty mice, which served as control were painted intravaginally twice weekly with acetone alone, and these animals revealed no evidence of malignancy. During the course of this experiment 28 mice died from some other diseases (no evidence of carcinoma). They were sacrificed on the 20th week after the first painting and observed microscopically. Malignant lesions were observed in 35 of the 69 C₃H mice (50.7%). Gross advanced carcinoma (invasive carcinoma) was found in 15 mice (21.7%), microscopic invasive carcinoma in 8 (11.6%), while carcinoma *in situ* developed in 12 (17.4%). Histologic types were as follows: spinal cell type 7, transitional cell type 12, spindle cell type 3, muco-epidermoid type 1.

The problem as to whether carcinoma *in situ* should be included in carcinoma remains open for discussion.

附 議

松井敬介：3・4-B.P. を用いてマウスに実験的に子宮頸癌を発生せしめる研究は、1 昨年すでに私が中村の協力をえて発表しており、昨年中村が原著として発表している。

陰癌の発生率があまりにも高率であったとのことであるが、これは (1) 術者の手技の拙劣によることが多い。(2) 1% B.P. を用いたとのことであるが、これは高濃度すぎる。0.1~0.3% 以下が望ましい。

(3) 用いられた系統は C₃H は適当ではない。この系統は癌好発系として有名な系統であるので、癌の発生し難い系統を用いるべきである。

細川：(1) 陰につかないようにという努力を十分注意するために小耳鏡を用いた。

(2) C₃H マウスを用いたのは早期癌から、浸潤癌に至る過程を知ることができたらと思い癌の生じやすいマウスを用いた。

166. ECOLOGICAL APPROACHES TO THE CAUSE OF STOMACH AND LUNG CANCERS

TOKURO SATO (Institute of Public Health)

1) Salted foods (brine concentration: 10~30%) cause haemorrhage and irritation in the stomach membrane of animals because of osmotic pressure. Deep or shallow scars have been observed according to the size and nature of the foods and emptiness of the stomach. In the cancer district where the highest incidence had been recorded in Japan, about 10% among those eating highly brined foods died of cancer. If it is assumed that 10% will die of cancer among those influenced by the agent and the life span to be 70 years, about 60% of the population will be under the influence in a district of the mortality rate of 80 for 100,000 population. The habit prevailing in the greater part of the population will easily be recognized by the people in and out of the groups, if one asks the names of or the procedures for the production of foods, as people eating highly brined foods do not taste salt, because of insufficient mastication. Making this as the working hypothesis several districts in Japan have been surveyed. Very high and low rates in several districts, extraordinary sex ratio, remarkable differences in places situated nearby, and remarkable decrease in the rates have been observed to be related to the habit.*

This hypothesis has also been shown to be tenable in north Wales, slums in Oslo, Finland, Seeland of Holland, Austria, and among Occidental Jews. In the districts of very low incidence such as Java (Dungal), and Thailand (observation of the people in London, Satoo), they take little of salted foods. People in British Uganda suffered for a long time from salt deficiency (Davies). The fondness of salted foods of the Japanese, who had been brought up in Japan and living abroad, is also noteworthy.

*Bull. Inst. Publ. Health, 8, 10, 187 (1959).

2) People in Europe often smoke very dry cigarette in winter. This is associated with heating system of rooms and this tendency is still increasing in practically every city in Europe. On the contrary in Japan they do not warm their rooms and smoke mostly humid cigarette in winter. The condensing action of water vapour in the cigarette has been calculated that humid cigarette smoked in low temperatured room retains about 10 times water compared with dry one in dry and warm atmosphere. When labile free radicals (Cuzin: Lyons, Ingram) and reactive substances are taken into consideration, the toxicity of dry cigarette in dry and warm atmosphere might be explained. To see the influence of dryness of cigarette the next study was made. Five countries (Austria, Finland, U.K., Ireland, Japan) where cigarettes

are principally smoked have been chosen. The mortality rate from lung cancer for several years are compared with the calculated net consumption of cigarettes by males and females in 20 years. The nature of cigarettes in Austria (about twice that of U.K.) and Finland is the worst. Air pollution appears not to take an important part for the causation. The rapid increase in the rate in Austria, Finland and U.K. is interpreted from the aggravation of the nature of the cigarettes. The mortality rate in Ireland is one third of that in U.K. and that in Japan is the lowest. These differences are in accordance with the humidity of cigarettes which is associated with heating of rooms in winter. From the view point of the radicals, freshly retained tar by pipe smoking will be toxic to the tissues such as the tongue and the lips.

Smoke was trapped in buffer solutions and cystein was added at 0 time or one hour after. They were condensed using charcoal and applied to paperchromatography, which was developed with acidic butanol and examined using ultraviolet light, ninhydrin and sulphur tests. Remarkable changes were observed especially with dry cigarettes.

(A part of the work has been assisted by W.H.O. as a fellow and by the Chester Beatty Research Institute as a visiting scientist.)

附 議

滝沢延次郎：胃癌と胃炎が同じ地方に多いからといってもそれが同じ原因で起こったかどうかはいまいと思います、演者の御実験で胃炎ができただけではそれを胃癌とすぐ結び付けるのは無理ではないでしょうか。

佐藤：この実験だけではその点まで触れることができないが、Hamperl, 村上, 久留等の炎症あるいは潰瘍と癌の関係を見た点、所の皮注の実験等を考えることができる。長期実験では慢性的の炎症は見られるが、最終の結果を見るまでに至っていない。

宮地 徹：食塩水を反復して肉腫を形成する実験は食塩水のみでなく、果糖、蔗糖、ただの蒸留水でも行われ、それぞれ肉腫が発生していることを御注意願いたいと思います。

167. THE CARCINOGENIC ACTION OF PARA-BENZOQUINONE ON THE LUNG OF MICE BY EXPERIMENTAL INHALATION (II)

MASAYOSHI KANISAWA and GENSHIRO IDE

(Dept. of Path., School of Med., Chiba Univ.)

We reported in the General Congress of the last year that the development of carcinoma in the lung of A-strain mice caused by 6 hours inhalation of vapour of para-benzoquinone 6 times a week in a closed tank with a capacity of 150 l., in which 10 mg of para-benzoquinone were evaporated by heater, was found in 3 cases out of 47 (6.4%).

In this time an experiment was done, in which A-strain mice inhaled 10 mg of para-benzoquinone for 3 hours consecutively twice a day, namely 20 mg for 6 hours a day, and 6 times a week during 200 days. And afterwards the action of the inhalation was discontinued and the animals were fed without any treatment in the same condition, as the control group.

The survived animals after discontinuance of the inhalation were 24 out of 30 in the experimental group and 26 out of 30 in the control group. 16 mice of them were killed on the 311th and 425th day after the first experimental treatment. The others died during the time.

The conspicuous finding was recognized in the lung of the survivals of the experimental group. The development of adenocarcinoma of the lung with extensive atypical proliferation of the bronchial epithelium or adenoma in other areas was found in 3 cases out of 4 which were killed on the 425th day. It proved to be 10% of the whole experimental mice. In the control group such a malignant tumor was not found, but only 2 typical adenomas and 1 adenomatous atypical growth were recognized. Furthermore, in addition to the carcinoma cases there were 3 cases (10%) which had adenomatous heterotopographical epithelial cell proliferation in the alveolar wall, and other 6 cases (20%) of adenoma or papilloma which showed more atypical feature than adenomas in the control group. They might be called malignant adenoma, for they showed the beginning feature of carcinoma.

From the above mentioned data it was established that the percentage of cancer development in this experiment was higher than that in the previous one, even if the difference of the experimental treatment was took into consideration, and that the development of the carcinoma in the lung of mice was found in spite of the discontinuance of the inhalation of para-benzoquinone.

附 議

宮地 徹: ベンゾキノン吸入による腫瘍で肺癌と呼んでられるのは、転移がみられたでしょうか。それがなければ癌とよぶのに慎重でなければならないと思いますが。

蟹沢：実験発生腫瘍が悪性腫瘍であるか否かには慎重であるべきだと私も考えております。従いましてわれわれの実験でも、生じた腫瘍が、破壊的増殖を示している場合にのみ、これを癌といたしております。また腺腫でその一部に形態学的にも、組織学的にも癌性の増殖を示す部分がありましたがこれらは緒方先生のいう癌の幼芽と考えて、対照例に見られる定型的腺腫と区別する意味で、仮に悪性腺腫という表現で示しましたが癌例には含めませんでした。なお転移は、悪性腫瘍の特徴ではありますが、一般に動物では転移が起りにくいとされております。

168. STUDIES ON THE HISTOGENESIS OF LUNG TUMOR INDUCED BY HYDRAZID

AKIRA YASUNO (Dept. of Med. Zoology, Showa Med. College)

The author conducted various investigations on the lung tumors induced in mice by oral administration of 0.25 per cent isonicotinic acid hydrazid (INH) histogenetically. The following results were thus obtained.

- (1) Generally speaking, most of tumors were found on the pleural surface.
- (2) Inflammation and atelectasis showed no particular causative relations with the formations.
- (3) The majority of the lung tumors (adenoma) induced by administration of INH per os were considered to be alveolar epithelial origin.
- (4) Malignancy could not be confirmed in the present experiment up to 18th month.

(文部省科学研究費による)

169. THE ELECTRONMICROSCOPICAL STUDY OF EXPERIMENTAL PULMONARY TUMOR IN MICE

(I) GENERAL CELLULAR REACTION IN ALVEOLI BEFORE TUMOR FORMATION

HAJIME KITAMURA and HIROYUKI SHINDO

(Dept. of Path., Nara Med. College)

There are two types of alveolar lining epithelium, but despite of works by many investigators the histogenesis of pulmonary tumor in mice is uncertain in the mean of above two cell types. The appearance of "hyperplastic foci" of alveolar cells was discussed by Simkin in the stage of pre-tumor-formation. In this study, the electronmicroscopical observation was examined about the general cell reaction of alveolar wall and especially so called "hyperplastic foci" of alveoli which appeared in the stage of pre-tumor-formation. The dd male mice were used, and group 1

were fed with 0.1% urethan in drinking water, group 2 were fed with 0.2% INAH, and group 3 were inoculated methylcholanthrene solution (0.1 mg each mouse) in the tail vein. Group 1 were observed at the 7th and the 10th week, group 2 at the 20th and the 30th week, and group 3 at the 6th weeks.

1) Alveolar epithelium: There was no proliferation of alveolar lining epithelial cells which were one type lacking osminophilic bodies and microvilli. But, the proliferation of another type of alveolar epithelial cells with osminophilic bodies and microvilli was found in the area of hyperplastic foci which were seen in the corner of alveolar wall. The decrease in osminophilic bodies and the increase in microbody or mitochondria and endoplasmic reticulum were found in proliferative alveolar epithelial cells of this type. The hyperlasia of this type of cells was observed more frequent in group 1 than in group 2. Usually, in group 2 and 3, the spindle shaped crystal bodies were found in the cytoplasm of alveolar epithelial cells with osminophilic bodies and microvilli.

2) Alveolar septal cell (interstitial cell): The proliferation of this cell was found in the area of hyperplastic foci, and there was the increase in cytoplasmic organella and vacuole, but interstitial elastic fiber and reticulum fiber were not so proliferative in hyperplastic foci.

3) Capillary endothelium: Nuclei of capillary endothelium were swollen and vacuoli appeared in its cytoplasm, but the lumen of it was narrow in the area of hyperplastic foci.

170. EXPERIMENTAL STUDIES OF LUNG TUMOR IN RAT, GIVEN CARCINOGENIC HYDROCARBONS IN COMBINATION WITH SEVERAL DUST OF MATERIALS

KAZUO TAKEMOTO

(Dept. of Public Health School of Med., Tokyo Med. & Dental College.)

For the purpose of experimental induction of pulmonary cancer, albino rats (Saitama strain) were given single intratracheal infusion of emulsified carcinogenic hydrocarbons (3 mg of 20-methylcholanthrene (MC) or 3, 4-benzpyrene (BP)) combined with one of the following several kinds of metallic and non-metallic materials. They were chemically insoluble particles of about 5μ of NiO (3 mg), Cr_2O_3 (20 mg) and Al_2O_3 (20 mg) and C (3 mg), BeO (20 mg) and SiO_2 (20 mg).

The table below shows the frequency in which tumors developed in this experi-

mental series. All neoplastic lesions were epithelial and all, except one adenocarcinoma induced by BeO were squamous carcinoma. There was no anaplastic carcinoma. No metastasis has yet occurred. BP was far less potent than MC in the induction of malignant lesions, although metaplastic proliferation occurred in the loci.

The process of tumor development followed the 4 morphological stages 1) non-motypical proliferation of cylindric epithelium *in loco*, 2) squamous and adenomatous metaplastic proliferation, 3) hyperplastic proliferation without atypism or infiltration, 4) atypical and infiltrative (neoplastic) proliferation.

As the matrix of neoplastic proliferation both bronchial and alveolar epithelia should apparently be considered. It was also noticed that squamous carcinoma often developed in the foci of adenomatous proliferation.

MC and BP were demonstrated to remain *in loci* by ultraviolet examination as long as 12 months. This finding, although contravessial to data stated in the literature, may be significant to explain the carcinogenesis.

Process of Tumor Development.

	1 m	2 m	3 m	6 m	12 m/	Tumor/Total	Malig.
NM	3(1)	5(4)	4(3)	8(6)	/	22/30	14
BM	0	0	3(1)	6(2)	8(5)	17/55	8
CM	0	0	2(1)	2(2)	/	4/40	3
SM	1(0)	0	1(1)	4(2)	/	6/40	3
AM	0	0	1(0)	1(0)	/	2/20	0
KM	0	0	0	1(0)	0	1/40	0
BB	0	0	0	0	0	0/25	0
CB	0	0	0	0	0	0/25	0
B	0	0	0	0	2(1)	2/70	1
C	0	0	0	0	/	0/20	0

171. PRIMARY SITE AND INVASION-FORM OF LUNG CANCER USING LARGE SECTION SPECIMENS

NOBUAKI KAMBARA, TORU MIYAJI, RYUHEI TATEISHI,
MOTOYOSHI SASAKI, KOJI WATANABE, TAKAYUKI YAMAUCHI
and GYOJI YANABU

(1st Dept. of Path. and Institute for Cancer Research, Med. School, Osaka Univ.)

On 168 autopsy cases of lung cancer we prepared serial large section specimens of entire lung mounted on the paper (by J. Gough and J. E. Wentworth), and on the other hand investigated the opposite portion microscopically.

By above mentioned method, we observed the spread form of primary site comparing the large section specimens with X-ray film in terminal stage, if possible, in early stage, and tried on the new classification for gross appearance of lung cancer.

RESULT:

Site of main tumor; 29.2% were peripheral, 35.1% intermediate, 30.4% hilar and 5.3% disseminated. Macroscopical type; 59.5% were massive, 15.5% fine nodular, 14.9% pleural and 10.1% peribronchial (details in table).

On the relations of the site and histological type, adenocarcinoma and anaplastic carcinoma were more often peripheral and hilar. Epidermoid carcinoma were more frequently intermediate. About the spreading form, epidermoid carcinoma and anaplastic carcinoma were more often massive and adenocarcinoma fine nodular (diffuse). Cavity formation and peribronchial invasions were seen more frequently in epidermoid carcinoma, and nodular dissemination of the same side in adenocarcinoma. Pleural invasions were seen equally in three types of carcinoma. Average survival duration was 10 months without relation to the site, and according to the macroscopical type that of the massive type (10.9 m) and pleural type (12.2 m) were different from peribronchial type (5.7 m) and fine nodular type (7.0 m).

The carcinoma with adjoining scar tissue (including tuberculosis) were found in 26 cases (15.5%). About the spread of the same side, pleural invasion were observed in 42.9%, nodular dissemination in 26.2%, metastasis of hilar lymphnode in 92.9%, pneumonic dissemination in 8.3% and pleuritis carcinomatosa in 8.9%.

Table

Site	Type	Number
Peripheral	Massive	15
	Pleural	25
	Fine Nodular	9
Intermediate	Massive	49
	Peribronchial	6
	Fine Nodular	4
Hilar	Massive	35
	Peribronchial	11
	Fine Nodular	5
Disseminated	Massive	1
	Fine Nodular	3
	Diffuse	5

172. THE SIGNIFICANCE OF SCAR IN THE DEVELOPMENT OF PERIPHERAL PULMONARY CARCINOMA

SHOICHI OBOSHI (Dept. of Path., School of Med., Hirosaki Univ.)

Out of 13 cases of primary pulmonary cancers recently experienced in our laboratory, 6 belonged to peripheral carcinoma, 1 to hilar carcinoma and the other 6 cases were too widespread to determine the primary sites. All of six cases of peripheral carcinomas showed a closed relationship to pre-existing scar tissue in their primary sites. But, no relationship to scar was seen in the peripheral zone of their primary tumors or metastatic growths. Accordingly, all of these cases seems to belong to the so-called scar cancer. The causation of the scars was tuberculous in 5 out of 6 cases.

The above findings seem to indicate that the majority of the pulmonary cancers may be originally developed in the pre-existing scar of the peripheral region of the lung. The significance of the scar as the regional disposition of the development of cancer should be appraised, as well as that of the carcinogenic substances as the exogenic causative factor.

(文部省科学研究費による)

附 議

今井 環：肺のはんこんを基地として、相当多くの肺癌が発生するという事は、きようであるかも知れないが、あるていど以上の大きさになった腫瘍では、たとえその部にはんこんがあっても、それが癌腫の母地になったかどうかを決める組織学的判定基準については、相当慎重でなければならないと思う。Rös-sle や Friedrich の基準だけであれば、ほとんどすべて、あるいは多数例の肋膜下癌に見られるという所見を、われわれは得ておる。癌塊の中心部がはんこん様になるのは、肺癌以外の癌でも普通である。したがって、はんこん癌に対する判定基準を、まずはっきり提示することが、必要ではないかと思う。

北村四郎：レスレーの癌痕癌に対する条件はもちろん重要な所見であります。さらに腫瘍以外の部の肺組織に腫瘍の間質と同性状の陳旧性炎があるかどうかを見る必要があると思います。またできれば慢性の刺激によって肺胞上皮なり気管支上皮が異型増殖を示すところから腫瘍への移行なり種々相を示すところが証明されることが望ましい。また初期癌の場合癌痕組織が腫瘍よりもさらに一層外側にも存在することが証明されるとよいと思います。

竹本和夫：1. 肺内癌痕と原発巣の関係として、そのでき方に、結核とともに珪肺性の線維増加を考慮に入れる必要があろう。われわれの 110 剖検例から、8 例の珪肺症と思われるものがあり、これも結核性線維ときわめて類似する故、判定には十分なる注意を用する。癌痕のでき方に偏光法を用いて炭粉等の粉塵を検する必要があろう。われわれの 110 例中癌痕癌は 16 例にすぎなかった。

2. 原発巣内に Elasto-fibrosis があるから癌痕癌という理論に対して、転移巣中心にも同様所見がある場合があり一言にはいい切れない。

大星：(1) 今井君に：癌痕が発癌以前に形成されたものであることは、その中に認められる陳旧性結核巣および高度の炭粉沈着で十分であると考えています。癌の中心部癌痕化ではこのようにはならないと考えられます。

(2) 北村君に：お話しのいろいろの点につきましては私どもも検討をいたしております。ただ、癌の中心部の癌痕の中に癌に移行する上皮の変化が見られねばならぬという御見解に対しては、私どもは出

来上った癌の中ではそのような移行はないと思っています。

(3) 竹本君に：確肺結節の可能性については検討致します。

(4) 家森君に：御教示有難うございました。

宮地：胃癌の術後1カ月で肺癌がみられたという第1例は、お話しになった組織像は燕麦細胞癌ではないと思います。粘液染色をおこなわれて、胃癌の転移ではないかという点をもういちど御検討頂けたらと存じます。

173. A CASE OF LUNG CANCER ASSOCIATED WITH ASBESTOSIS

NOBUAKI KAMBARA, KENJI NASU, SHUNZO ONISHI,
MOTOYOSHI SASAKI and TORU MIYAJI

(1st Dept. of Path. and Institute for Cancer Research, Med. School, Osaka Univ.)

A 65 year-old male, who died in December 1959, had been working in an asbestos-factory as a fiberizer for about 35 years from 1918 to 1953.

In 1946 and 1949, he was diagnosed as chronic bronchitis and pulmonary tuberculosis respectively.

In 1953, by mass examination, he was diagnosed as an asbestosis at the Osaka Koseien Sanatorium. At that time, the X-ray film of the chest revealed many fine nodular and streaky shadows in all area of both lungs, mainly middle and lower portion, associated with pleural thickenings. Smears and cultures of sputum for tuberculous bacilli were negative.

Before two months of his death, a homogenous shadow was first recognized roentgenologically in the left upper area near mediastinum, and cancer cells were also noted in sputum.

Autopsy disclosed a bronchial carcinoma of the size of a chicken-egg, originated in upper middle area of the left lung, which revealed epidermoid carcinoma partially with adenomatous pattern histologically. Metastases were found in the both lungs, both kidneys, pancreas, liver, ribs, vertebral spines and bilateral hilar lymph-nodes.

Besides the carcinoma, there were diffuse fibrosis with many depositions of asbestos bodies, especially in alveolar, perivascular and peribronchial areas. Other findings were cuboidal cell metaplasia of alveolar cells and bronchiolar epithelia, micro-abscess in hilar region, emphysema of subpleural region and pleural thickenings, but squamous metaplasia of bronchial epithelium was not detectable.

VII. Virus

174. ELECTRON MICROSCOPIC OBSERVATION OF CHICKEN SARCOMA CAUSED BY INTRATHYMIC INOCULATION OF VIRUSES

TAMAKI IMAI, HIROMITSU OKANO and YŪKŌ NISHIUMI

(1st Dept. of Path., Faculty of Med., Kyushu Univ.)

The virus isolated from a chicken sarcoma of the Chiba strain was inoculated intrathymically in suspension in a dose of 0.5 cc into white leghorns by a method of the authors' own devise. An electron microscopic observation revealed the following findings:

4 days after inoculation: In the interlobular stroma of the thymus were seen an increasing number of tumor cells with virus particles attached to their surface. The virus particle was round or ovoid in shape, averaging 75 m μ in diameter, with a nucleoid in its center and a single or double membrane surrounding it.

6-7 days after inoculation: Inspection of each single section of the tumor cell revealed in 3 out of 8 cases that some of the cells had each 200-500 virus particles deposited in it and others had viruses arranged almost uninterruptedly on their surface; that the cytoplasm was in part occupied by a space bounded by a limiting membrane, complex in shape, and filled with innumerable viruses and some electron dense material; that the cytoplasm with such a space in it underwent degeneration, with disorganized and vesiculated endoplasmic reticula, a reduced number of Palade's granules, and vacuolated mitochondria occurring there, and was frequently in process of disintegration round about the more compact of the aggregates of viruses present in it; and that the virus particles in the cytoplasm, those in an intracytoplasmic space, in particular, were nonuniform in shape, structure, size and electron density.

200 γ X-ray irradiation of the tumors on the 8th day after the inoculation produced these changes in 24 hours: A change in the number, position and arrangement of viruses; disappearance of intracytoplasmic spaces, though the viruses were detectable in a little greater number in the irradiated than in the nonirradiated tumors; most of the virus particles were seen occurring in tiny cluster a trifle outside, and not inside, the cell membrane.

附 議

新保幸太郎: ① Virus の増殖は細胞膜の直上で起るか, 直下でおこるかについて承りたい。

㊦ Virus 粒子の多い細胞と少ない細胞では、その変性程度に著明な差異がみられますかについて承りたい。

今井：① 腫瘍細胞表面に多数のウイルス粒子が認められる場合でも、細胞膜の直下にはウイルスの増殖と関係あると思われる所見は見えておりません。

② ウイルス粒子の多数見られた腫瘍細胞では変性の像が見られることがあるが、ウイルス粒子の少ない細胞では細胞質の変化は少ない。

175. EXPERIMENTAL STUDIES ON HISTOGENESIS OF VIRAL TUMOR. (III) COMPARATIVE STUDIES ON THE THREE STRAINS OF CHICKEN SARCOMA

KOTARO SHIMPO, EIMEI NARIMATSU, HIROSHI HIGASHI,
KAZUMI NISHIDA and KOZO MUROYA

(Dept. of Path., Cancer Institute of Sapporo Med. College)

A comparative study was made on the Chiba-, Aichi-Eiken-, and Rous No. 1 strain of chicken sarcoma pathohistologically, and electron-microscopically.

In these three strains, when examined in detail, histological findings are more or less different from each other.

The latent period of the tumor growth was shortest for Rous No. 1 strain, and longest for Chiba strain, being parallel with the length of host survival time.

The Chiba strain was characterized by absence of metastasis, and by marked production of mucinous substances. Light microscopically, spindleshaped tumor cells were found to be predominant cellular constituents of the tumor, whereas tumor cells of the other strains tend to show a certain pleomorphism and degeneration. Electron-microscopically, distension of E.R. with Palade granules was distinctly observed. Electron-microscopical detectability of the virus particles was lowest (0.32%) among three strains, however increased remarkably by X-ray irradiation (36.0%).

In Aichi-Eiken strain, on the other hand, a high incidence of metastasis in the lung, liver, and heart, and hemorrhage into the tumor tissues were noteworthy. The tumor cells were pleomorphic and giant cells were seen frequently. Electron microscopically, one of the most characteristic findings was a marked development of Golgi apparatus. The detectability of the virus particles was 5.14%. This detectability was increased about four times as much as that in control (22.3%), by X-ray irradiation.

The tumor cells of Rous No. 1 resembled those of Aichi-Eiken strain, but the detectability of the virus particles was much higher (10.3%) than in the other strain.

After X-ray irradiation, the detectability of virus particles was found to be highest among three strains (50.9%).

It was examined with Rous No. 1 strain, whether or not the increased virus particles in the irradiated tumor tissues are infective. By pock counting method, infectivity titer of the irradiated tumor was found to be twice as high as that of the non-treated control.

176. CHEMICAL STUDIES ON CHIBA STRAIN CHICK SARCOMA VIRUS

TAIJO TAKAHASHI, MINORU ISHIZAWA and TETSUYA TAKAHASHI

(Section of Chemistry, Research Institute of Cancer, Faculty of Med., Kyushu Univ.)

Our attempt is to research for tumorigenesis by following up the virus multiplication in fowl sarcoma cells. This is the preliminary result at the beginning of our investigation. At first, the virus was tried to be purified by fluorocarbon-extraction from the sarcoma inoculated in thymus of chick, which contained more viruses than in wings, or muscle. After fluorocarbon-extraction of 5-7 times, the aqueous layer containing virus was centrifuged at 105000 G, for 90 min. at 0°C. The infective pellet obtained above had high concentration of ribonucleic acid. Electron microscopic studies, however, showed this pellet contained the smaller particles than virus (80m μ). Therefore further centrifuging fraction was necessary for biological and chemical study. Secondly, the homogenate of tumor treated with hyaluronidase was centrifuged repeatedly at 105000 G, for 90 min. at 0°C. The fourth supernatant which was taken by pipett from the top layer of the fluid in every centrifugation had still infective potency. This suggests the possibility that the initial homogenate has high infective titer or that the very small granular or soluble infective materials exist in it. Further study is under way on this point.

177. HISTOLOGICAL & ELECTRONMICROSCOPICAL STUDIES OF FRIEND DISEASE

TSUTOMU KASUGA, YASUHIKO SHIRASU and HIROYASU MIYAMOTO

(Dept. of Path., Cancer Institute)

Correlated histological, hematological and electronmicroscopical studies of Friend's disease were conducted in the hope of elucidating the nature of the disease in the

mice. *In toto* 70 Swiss strain mice were inoculated with intraperitoneal dosage of 10% homogenate of infected mouse spleen. The development of the lesions in the organs and change in the peripheral blood of the infected mice was followed up to eight weeks at regular intervals.

Remarkable splenomegaly, moderate lymphadenopathy and elevated nucleated cell counts in the peripheral blood were observed quite regularly. Each change reached its acme at the end of the third week. In the spleen and nodes, the initial lesion was invariably a reactive proliferation of the reticulum cells at the perifollicular marginal zones, starting as early as 24 hours post inoculation. This was followed by the involvement of the germinal centers. On the fourth day, groups of atypical cells with a hyperchromatic nucleus, conspicuous nucleolus and hyperbasophilic cytoplasm began to appear in the pulp, especially in the subcapsular areas. They grew rapidly until almost the entire structures were replaced by the atypical cell proliferation, although the fundamental architectures of the organ were well preserved.

Although the findings appear consistent with classical leukemic changes, there are still several points which may contradict with the interpretation. The elevation of the nucleated cell counts of the peripheral blood was mainly due to marked increase of erythroblasts and not to the presence of the atypical cells which, if present, did not make more than a few percent. The peripheral erythroblastosis was closely associated regularly with occurrence of foci of erythroblastosis in the midst of the areas of hyperplasia of the atypical cells in the spleen. Granulocytic series, especially the mature leucocytes, many megakaryocytes were observed in unusually dense population in the atypical cell foci. The bone-marrow was very rarely affected by the atypical cell proliferation, and even if there was any of such foci, these in the earlier stage, practically none of the marrow in the later stage showed any atypical cells. Although in about half of the mice the liver was affected in the later stage, the infiltrating cells suggested often autochthonous proliferation of the reticuloendothelial elements.

The atypical cells observed in the spleen and in the peripheral blood were negative for nadi-oxidase reaction. Electronmicroscopical features of the cells include abundance of RNA granules, scarcity of the organelles, especially of the endoplasmic reticulum, and suggested close similarity of the cells with the reticulum cells. Type A viral particles of Bernhard, with the outer shell diameter of 790 A & the core of 460 A, were observed on the cell membranes and in the endoplasmic reticulum of the atypical cells. Although no definite inclusion body was found, an irregular accumulation of RNA granules, in continuity to the endoplasmic reticulum which contained the viral particles, was noticed. The viral particles were also observed in the normal looking reticulum cells, although the megakaryocytes and erythroblasts

failed to do so.

It is concluded that in the Friend's disease the virus is definitely operative in inducing the characteristic lesions, that the atypical cells are most probably of the reticulum cell derivatives, and that there exist several points in the entire morphological features which prevent the disease entity from which should be elucidate being interpreted as quite comparable with classical leukemia.

附 議

天野重安: 1. ただいまの大型の細胞は炭粉そのたコロイド粒子を貪食する能力をもっていますか。

2. 骨髓の所見が、一過性または転移巣を作るのみであるというように承ったが、それでは白血性になっていないわけですが、白血球数の最高値になった場合およびその際の例の大型細胞のパーセントをお教えなう。

春日: ① 貪食能の有無についての実験は行っておりませんが、崩壊細胞成分および赤血球を取り込んでいる像は認めております。

② 一過性に小転移巣が認められたのではなく、2 週および3 週の各一例にやや強い異型のな細胞の増殖が認められたにすぎませんので、異型細胞が末梢血中に出ただけでは白血病とはいいい難い と思います。

③ 有核細胞数の最高値は 60 万でその例における大型異型細胞はほぼ数%程度でした。ただし裸核細胞は除外しております。

中原和郎: 重要な点であるが、Friend の leukemia ではスィスマウスを試験動物とすることを強調されている。日本の“スィスマウス”が果してその規格に適合しているかどうか分らない。したがってここで見られた病変が典型的な Friend leukemia といえない可能性がある。

釜洞: 中原氏のいわれたごとく Swiss adult mice に限定されるのが Friend virus の特徴だからこの点よくたしかめて頂きたい。

春日: 20 代以上経代された近交系 Swiss mice を使用しました。Friend virus は Friend 博士より送られたものです。

Swiss adult mice および DBA 12 mice のみしか Friend virus に感受性がないといわれておりましたがその後 Furth らは RF strain mice にも感受性ありと述べ、また最近白須、大橋は ddY 系マウスを使用して Friend 症候群を再現しております。これらの事実は系統差による感受性云々という問題について再び検討して見る必要があることを示しているものと存じます。

178. THE ISOLATION OF TOXOHORMONE FROM FRIEND VIRUS INFECTED MOUSE SPLEEN

MOCHIIHIKO OHASHI

(Cancer Institute, Japanese Foundation for Cancer Research)

The isolation of the liver catalase depressing factor, Toxohormone, from malignant tumors has been confirmed by many investigators. It has been shown that Toxohormone is contained in large amounts in all the malignant tissues so far studied.

The nature of the leukemia-like disease in mice which is transmissible by virus discovered by Dr. Friend, has been discussed from the pathological observations and the transplantability among the pathologists.

In this report, in order to elucidate the subject from biochemical viewpoint, we extracted Toxohormone from the huge spleen which was collected from the Friend virus leukemia mice three weeks after the virus inoculation, and assayed its catalase depressing activity. The result indicated the Toxohormone activity of the Friend leukemia spleen was fully as high as that of rhodamine fibrosarcoma.

179. DEVELOPMENT OF LYMPHATIC TUMOR IN DDO AND DD₁ MICE PRODUCED BY MOLONEY'S LEUKEMIA VIRUS, RECOVERY OF VIRUS AND SOME EVIDENCE OF VIRAL TRANSMISSION TO THE PROGENY

YOSHIHIKO TSUBURA, KUNIMICHI ICHIBA, IKUO KIMURA
and KYOICHIRO MATSUMOTO
(Dept. of Path., Nara Med. College)

Non strain specific leukemia virus was extracted from sarcoma 37 by J.B. Moloney. Lyophilized virus was obtained from his laboratory and tested in ddO mice which has high incidence in spontaneous mammary tumor but low in leukemia. When 0.1 cc of hundred fold diluted virus was inoculated into 14 mice of three month old intraperitoneally, 2 of them developed lymphatic tumor; 14% of incidence at an average survival of 189 days after the inoculation. When same amount of virus was inoculated into 46 newborn mice, lymphatic tumor occurred in 20 mice; 48% of incidence at an average survival of 223 days. Incidence in females was 52% with an average survival of 214 days and that in males was 41% with an average survival of 239 days. Storage of hundred fold diluted virus at -20°C decreased the incidence to 25%. Virus recovered from the leukemia virus induced tumor tissue was tested its activity in dd₁ strain of mice which is high in mammary tumor incidence but low in leukemia. During 117 days under observation, incidence of lymphatic tumor was 44% at an average survival of 139 days.

Sixty-three mice, whose mother or father, or both of them were inoculated with 0.1 cc of leukemia virus, did not develop tumor within 200 days of age. A 76-day-old male mouse, whose mother received intravenous inoculation of virus one day prior her delivery, died with enlarged spleen, weighing 535 mg. 10 mg of spleen, which revealed no evidence of tumor histologically, was grafted subcutaneously into fifteen of 55 day old mice whose parent were received the inoculation of virus 23 days before the child-birth, and three died of lymphatic tumor at the ages of 135, 149 and 197 days. This result suggests that leukemia virus is transmitted from the parent and propagated in the progeny in these conditions.

附 議

入野昭三：胸腺のいちじるしい腫大を示す型と然らざる型との間に末梢血液像，主要臓器への細胞浸潤様式などに関して差はないでしょうか。私達は線照射によって惹起せしめた RF 系マウス白血病についていわゆる胸腺腫型と非胸腺腫の二型のあることを指摘し，両者間に明らかな相違のあることを認めましたのでお尋ねいたします。

螺良：岡山大学の報告を見まして，こちらでも脾臓，胸腺，リンパ節の大きさと血中白血球数の相関をしらべましたが相関関係はないと思われます。

浜崎幸雄：Polymer や Rous V でも Virus が自然感染した場合には Tumor を作らない。したがって演者の動物にリンパ腺腫がなかったから Virus の自然感染がなかったとはいえない。

螺良：この場合感染していないということは，発症しないからといっていえないことはもちろんで，この腫瘍化していない脾臓も感染していると考えられることもできると思います。ただ私の実験の判定の基準は発症するか，しないかを基準にしたのであります。

釜洞：1. フレンドとモロニーによる腫瘍は形態学的に区別できますか。

2. この仕事の重点ならびに今後の方向はどの辺にあるのでしょうか，遺伝的な問題でしょうか？

螺良：遠心分離だけではわけにくいと思いますが，生物学的には Friend はスイス系にのみ感受性があるという点で区別できると思われます。

相沢 幹：釜洞座長に：Polyoma virus と Friend virus については，免疫学的な同定の方法がある。

演者に対し：contamination については，発症という立場と，発症のない抗体形成の検索も必要でしょう。

180. PATHOLOGICAL STUDY OF MOLONEY'S LEUKEMIA VIRUS INOCULATED MICE

YOSHIHIKO TSUBURA, HAJIME KITAMURA, KUNIMICHI ICHIBA,
IKUO KIMURA, MASAO MARUGAMI, KYOICHIRO MATSUMOTO,
and MASAO NAKAZAWA (Dept. of Path., Nara Med. College)

ddO mice were inoculated with 0.1cc of hundred fold dilution of lyophilized Moloney virus intraperitoneally when newborn, and tumor occurred in 20 mice at an average survival of 223 days. Grossly main involvement of tumor was in spleen, thymus and lymphnodes. Thirteen cases showed remarkable enlargement of thymus obscuring the heart and lung in the pleural cavity. The rest of the cases revealed enlargement of spleen or lymphnodes without involving the thymus. Lung, liver, kidney, parotid gland, testis or ovary and pancreas were also infiltrated with tumor cells. Involvement of bone-marrow was seen in 2 cases. All the inoculated mice were counted the number of blood cells obtained from the tail vein fortnightly. No remarkable change was seen in the number of white or red blood cells except for some of tumor mice in their late stage. Histologically, the tumor cell was larger than lymphocytes, sometimes resembling to a reticulum cell without forming reticulin fiber. According to the locomotion of living tumor cells observed under

phase-microscopy and the results of May-Gruenwald staining and peroxidase reaction, the tumor cells belonged to the lymphocytic origin. In the early stage of involvement, the tumor cells were seen in the center of lymphfollicle in thymus, spleen or lymphnode, then proliferated to spread out the surrounding tissue. At later stage, the tumor cell became rather small, round and uniform in shape representing moderate mitotic figures. These findings led to the conclusion that the tumor was lymphosarcoma originating from the center of the lymphfollicle. No other kind of tumor was observed in tumor mice. Electron-microscopy revealed virus-like particles in an extracellular or intracytoplasmic position. Particles were less in number in thymus or lymphnodes compared with those in spleen.

181. ELECTRON-MICROSCOPIC STUDIES ON MAMMARY CARCINOMA OF THE C₃H, SL AND DDN STRAINS MICE, WITH ESPECIAL REFERENCE TO PROLIFERATION MODE OF VIRUS PARTICLES

KEISUKE MATSUI, SHOSUKE MORIWAKI
and KENTARO WATANABE
(Dept. of Path., School of Med., Tottori Univ.)

Each three cases of spontaneous mammary carcinoma (adenocarcinoma) of the C₃H, SI and ddN strains mice were examined electron-microscopically. Proliferating mode of virus particles was especially investigated.

Results obtained are as follows:

1. Mature viruses having nucleoid body are observed mainly within the duct lumen or intercellular spaces, but a few of them are observed in the cytoplasmic ground substance of the cancer cells.
2. Virus particles are proliferating from the cancer cells (a) at the top of the microvilli facing to the duct-lumens, or intercellular spaces (b) at the cellular surface, (c) at the matrix-like body, and (d) around the limiting membrane of the intercellular ductless-like spaces.
3. Proliferating modes from the top of the microvilli and the cellular surface are divided into two types, developing with nucleoid body, and without it (seldom).
4. There seems to be seen relationship between above mentioned four types of proliferation modes.
5. For a proliferation of cancer viruses, a nucleus of the cancer cell is not seen to be more important than cytoplasm of it.

附 議

天野重安：松井教授へ：貴検索の C_3H 、マウス乳癌材料が細胞表面と細胞質基質内と二種のウイルス増殖様式を示していることについてはわれわれも賛成し得ますが、SL マウスではわれわれの場合は細胞表面増殖のみでした。貴例の ddN ではこの二様式が現われていますか伺います。

松井：検索された 3 系統、すなわち C_3H 系、SL 系および ddN 系すべてのマウス乳癌細胞に、細胞表面と細胞質基質内との 2 種類のウイルス増殖形式が認められました。細胞表面増殖形がどの系統のマウスにも随所に多数認められるのに比して、細胞質基質内増殖形は一般に少なく、それでも C_3H 系にはしばしば見られますが、SL 系ではときに観察される程度、ddN 系ではまだごくわずかしき観察されていません。しかしながら、この細胞内増殖形の存在は、 C_3H 系マウス乳癌では文献上の記載もいくつかあり、しかもこれが従来もっとも注目されていた増殖形式であることは周知のとおりです。ただこれがいかなる経過をたどって細胞外に排泄され、そこで核様体を有する成熟ウイルスになるのかということについて、従来の報告では納得できませんが、contamination と解釈することも妥当ではないと思われます。今後この点を追求する必要があると考えています。

釜洞醇太郎：Bernhard の A, B 等の粒子についての先生の御見解をおたずねしたいと思います。

天野重安：最初から癌ウイルスは細胞表面増殖を呈するという枠があったのではなく、観察の要約が、たまたまわれわれが SL マウス乳癌を材料とした関係でそういわせるようになり、かつ、同時に観察した他の肉腫や白血病（マウス、ニワトリ）がこれを他面から支持するようになったわけです。

SL マウスの白血病ウイルスは白血病嫌発性の S マウスに接種することで、S マウスに白血病を相当率発生させることに成功したので、このウイルスの原因体としての資格は確認されたわけです。

また、Moore 等は C_3H 乳癌細胞の培養で、表面増殖ウイルスと細胞質内の小型ウイルスとの間に直接移行像のないことを認めている。その後米国ではポリオーマウイルスの汚染などで種々ウイルスの同時感染の案じられる所見がでてきているので、これからは、特別の操作で検討の必要があるように考えています。

182. ON THE ACCELERATION OF BREAST CANCER OF C_3H MOUSE COUSED BY THE AGENT ISOLATED FROM LEUKEMIC AKR MICE

YUKIO HAMAZAKI and MOTOMASA MURAKAMI

(Dept. of Path., Med. School, Okayama Univ.)

Previously, we have succeeded in isolating the filtrable agent from leukemic AKR mice, and the agent has been inoculated into the brain of C_3H newborns less than 12 hours old. The newborns of the same parents have been divided into two groups, i.e., the test group and the controls. Of 106 test group inoculated with the agent, 2 have developed leukemia and 15, mammary cancers. Of 83 controls inoculated with the heated agent, 9 have developed mammary cancers. That is to say, the number of mammary cancers of the test group may be superior to that of the controls at the rate of 5 : 3. Histologically, in the test group, mammary cancers have been constructed from the compact masses of round cells in most instances, and sometimes showed anaplastic glandular types with more mitoses. Metastasis has

occurred to the lung, but not to other sites. Generally, the infiltration of lymphatic cells and the proliferation of mesenchymal cells have been observed in alveolar septums of the lung, where the septums have become thick. As to the liver, lymphatic cells have infiltrated into the sinusoids, besides small groups consisting of some stellate cells and a few monocytes have been recognized, while in the spleen giants cells as well as reticular cells have proliferated in red pulp. On the contrary, in the controls, mammary cancers have proved to be cystcarcinoma haemorrhagicum. But we have not encountered the histological changes in the other organs of the test animals developed mammary cancers.

We have investigated sero-immunologically the correlation between the filtrable agents from virus tumors in this serial experiment concerned, and it has been confirmed that the agents have commonly had an antigenetic factor, however an identical antigenicity has not been between the mammary cancer agent of the test group and normal C₃H.

(文部省科学研究費による)

184. CLINICAL STUDIES EMPLOYING THE ONCOLYTIC VIRUS "E.D." (IV). CONCERNING THE THERAPY ABOUT HEPATOMA

- (1) MASAEI MIYAZAWA, MASAO NISHIKAWA, TERUO USHIO,
SHIGEO MATSUMURA, KIYOSHI SUGIYAMA and AKIRA UENO
- (2) TADASHI YAMAMOTO, KUSUYA NISHIOKA and AKIRA ODA
- (3) HAYAMI KINUGAWA

- (1) Surgical Dept., Okubo Municipal Hosp.
- (2) Oncolytical Dept., Institute for Infectious Disease
- (3) Dept., of Hygi., Chiba Univ.

In the 3rd symposium of cancer research at Nagoya the authors reported a case of metastatic hepatoma which was markedly improved by the direct perfusion of E.D. virus into portal vein and aorta descendens. Experimental studies confirmed the effectiveness of the periliental perfusion of E.D. virus against the metastatic hepatoma C₃H mice which were previously transplanted translienally with MH 134 ascites hepatoma cells. The survival rate of the treated mice were markedly increased especially in the group of mice perfused 48 hours after transplantation. No or scarce increase was observed by the other route of treatments. Distinct necrosis of MH 134 hepatoma cells in the liver was also observed histologically.

CLINICAL: On the other hand clinical cases of primary hepatoma and metastatic hepatoma were treated by the direct perfusion of E.D. virus.

CASE 1. ICHIKI, female, 63 aged. Metastatic hepatoma recurred from cancer of colon desc. resected 6 month ago. The liver as a whole, especially in left lobus,

was nodulously hard, swollen by a handbreadth under the left costal margin. E.D. virus preparation was perfused in ca. 10 cc from portal vein and in ca 13 cc from aorta descendens. After 10 days the tumor reduced amazingly and became much softer. Lethal 2.5 months later.

CASE 2. MIZUNO, male, 46 aged. Primary hepatoma. The left lobus of liver was small-nodulously hard, swollen and protruded by half handbreadth beneath the left costal margin. 15 cc of E.D. virus preparation was perfused into the supermesenterial vein. After 23 days the liver surface sank from the costal margin and mach softened. Another 12 cc of E.D. virus preparation were perfused from femoral vein. Generally improved and left the hospital. Another primary liver cancer and two metastatic hepatomas are omitted here to refer in detail. The immune responses of all cases were traced but the HI and CF titers were recognized to be not so high and very slowly increased than in the previous cases treated intravenously. Histological observations also confirmed the distinct effect.

VIII. Diagnosis

185. STUDIES ON MALIGNOLIPIN

TAKEKAZU KÔSAKI, NAYAO UEZUMI, SHIN'ICHI HASEGAWA,
SHIN'YA NAKAGAWA, YOSHIMARO KOTANI and KEISHI MURAKI

(Dept. of Biochem., School of Med., Mie Pref. Univ.)

Kôsaiki and co-workers isolated, three years ago, from human malignant tumors a new phospholipid, malignolipin, composed of spermine, choline, phosphoric acid, and fatty acid. With the paper-chromatographic method for the detection of malignolipin in blood and ascites, blood of 1524 human subjects as well 453 experimental animals and ascitic fluid of 365 experimental animals were examined. Malignolipin could not be detected in blood of normal mice and rats, but could always be detected in the blood of rats and mice bearing fully grown malignant tumors, and also in tumor ascitic fluid of rats and mice. Malignolipin appeared in the blood as early as 1-3 days after the inoculation of malignant tumors. Malignolipin was detected in the blood of all 465 subjects bearing histologically proved malignant tumor including diverse types of tumors, but never in the blood of all 493 subjects highly exclusive of malignancy, including 363 patients suffering various kinds of diseases, 90 normal subjects and 40 pregnant women. A method to isolate malignolipin as its picrate, and to obtain its ammonium salt removing picric acid was reported. Malignolipin picrate is stable enough. A method to detect and measure malignolipin contained in tissues as its picrate on the same principle was devised, and it was ascertained with this method, that malignolipin could always be found in various tissues of tumor bearing animals, but never in tissues of normal animals, including embryonal or regenerating tissues. It was then demonstrated, that the subcutaneous injection of an ammonium salt of malignolipin resulted in the production of a complement-combining specific anti-body in rat's sera, which was not pre-existent in normal rat sera. Further, the anti-malignolipin rat serum was ascertained to exert a considerable effect on the vitality of homologous Yoshida sarcoma cells, although normal rat sera caused no change.

附 議

原 義雄：本反応の追試成績については、昨年の本学会で報告したが、その後症例を加え、癌 219 例中 185 例 (85.4%) 陽性、非癌 412 例中 367 例、すなわち 86.6% 陰性で、これまで追試したいろいろな癌反応中、もっとも適中率が高かった。

徳岡俊次：Malignolipin の構成成分といわれました Spermine に関しては、私はすでに 11 年前に癌組織

および癌患者血清中に Spermine が増量していることを初めて確証したことを発表し、同時に血清スベルミン反応を提唱し、その後本学会でも発表しました。いままで 3200 余例の悪性腫瘍患者で、適中率 85% 内外の成績を得ています。しかしテンカン、脳腫瘍、妊娠中期に 20% 近く陽性を示すことがあり、また肝や腎の機能障害のあるものや肺結核の重症例でも 5~10% 陽性に出ます。しかしスベルミン反応は今日世に提唱された癌反応中では群を抜いて実施が簡単ですから、集団検診への応用に適していると思います。本邦でもすでに実施されているようです。この際本反応疑陽性のものには Malignolipin の検索を行うようにすれば癌の集団検診に大変役立つのではないかと提案します。

森下宗司：われわれも産婦人科に関する諸疾患について、検査してみましたところ始めて間もないのかかわらず 93.8% という癌適中率を得ました。

follow up をしていないので「ビラン」「内膜炎」の陽性はなんともいえません。妊娠に高率に偽陽性がありますが、神前氏のいわれるスポットの形等に注意する必要があると思います。妊娠の 4~5 月頃は最も代謝が異常を来すので「ニンヒドリン」陽性物質が同じ辺に出現することはあり得ると考える。このために今回の演者のビクラート法を併用したならば、ことに妊娠を除外すれば臨床的に使用する価値のあることと思います。

186. AN EVALUATION FOR MALIGNOLIPIN TEST

SHÔITI YAMAGATA and ICHIRO NAKAGAWA

(Yamagata's Clinic, Med. School, Tohoku Univ.)

1. We tried malignolipin test for the patients hospitalized into our clinic, totally 173 cases.

In 94 cases diagnosed as cancerous or malignat disease, 95.7 per cent was positive, and in 79 non cancerous cases 39.2 per cent was positive, then we could not recognize this test as cancer specific reaction.

2. We recognized that ^{32}P was incorporated into malignolipin and so called spermin fraction, extracted from Ehrlich ascites tumor cells, and liver, kidney of host animals by using of macroautoradiography.

3. When 100 μc of $\text{Na}_2\text{H}^{32}\text{PO}_4$ diluted with physiological saline solution was injected into the abdominal cavity of dd-mouse, we recognized that ^{32}P incorporated into malignolipin fraction was maximum in 48 hours after injection.

4. We recognized that ^{32}P incorporation into malignolipin fraction was maximum in 3 hours after injection, when the mixture containing 2 cc of ascites and 1 cc of YLA medium, using of roller tube method, was incubated at 37°C , but ^{32}P incorporation into phospholipid fraction, by Thanhauser-Schmidt method, increased even in 9 hours after injection.

Reference

- 1) Takekazu Kôsaki; Science, 127, 1176 (1958).
- 2) M. Friedkin, A.L. Lehninger; J.B.C. 177, 775 (1949).
- 3) C.G. Huggins, D.V. Cohn; J.B.C. Vol. 234, 257 (1959).

187. THE EVALUATION OF DETECTION OF MALIGNOLIPIN IN BLOOD IN THE DIAGNOSIS OF MALIGNANT TUMOR

MASAZUMI MIZOKUCHI and HARUMITSU KOBAYASHI

(2nd Dept. of Surgery, Faculty of Med., Kyushu Univ.)

The diagnostic value of detection of Malignolipin in blood (Kôzaki) for malignant tumors, especially for carcinoma of the stomach is discussed and clinically important findings are obtained.

The results of this test in 321 cases are carefully analysed. Even in the control group there are 18 positive out of 20 cases with hepatic impairment (B.S.P. more than 15%) and 6 positive out of 8 cases with renal lesion. It is obvious that for cases with hepatic or renal lesion we must be very critical in evaluating positive result. If these cases of hepatic or renal lesion are excluded there are 99.0% positive in 96 malignant cases and 15.5% positive in 181 control cases. Thus the diagnostic value of this test is sufficiently recognized.

In cases with gastric or duodenal lesion there are 88 positive out of 89 cases with carcinoma of the stomach (98.9%) except one case of mucosa cancer. It is interesting that there are 6 early carcinomas of the stomach in these 88 positive cases. While in 80 control cases with benign lesion of the stomach or duodenum there are 22.5% positive. Besides there is one positive out of 3 cases presenting pictures suggestive of precursor of cancer.

In conclusion, it is found that when this test is employed it is important to find out the presence of hepatic or renal impairment. Reevaluation of this test on the ground of our new finding may lead us to recognize this test as a significant mean of cancer detection.

188. STUDIES ON FLUORESCENT DYE (NTS), ITS AFFINITY FOR MALIGNANT TUMOR CELLS AND EFFECTS ON THE DEVELOPMENT OF TUMOR

SHOJI HATTORI, MINORU MATSUDA and KENJI MORIMOTO

(The 3rd Dept. of Int. Med. Osaka Univ.)

Newly discovered fluorescent dye (NTS) has a specific tendency to combine with basic protein *in vitro* in the alkaline solution more than at pH 6.3.

1. The dye was injected into tumor bearing mice intravenously. The mice were

sacrificed and macro- and microscopic findings were observed in 24 and 48 hours after injection. The affinity of the dye with tumor cells were marked in the site of chromatin and nuclei in the cells. Fluorescence was observed until two weeks after injection. However, the fluorescence was able to detect in some part of normal testis or caseous tuberculous lesions, therefore it seemed to be rather unspecific in the tumor cells.

2. NTS, No. 26 were injected intravenously into 11 cases of cancer, 1 case of gastric ulcer and 1 case of mediastinal dermoid cyst with a daily dosage of 50 and 100 mg per person for 2 to 6 days. Removed tumor revealed fluorescence except in the area of cancerous ulcers or the benign tumor cells. No side effect was noted in this series by intravenous injections.

3. Intraperitoneal injection of NTS, No. 26 and No. 23 into mice, which were inoculated Ehrlich or sarcoma 180 tumor cells subcutaneously, showed marked inhibition of the development of tumor with a daily dosage of 200 mcg per mouse for 7 and 19 days. Intravenous injection of 100 mcg per mouse for 7 days showed similar efficacy. Subcutaneous injection of 100 mcg per mouse also showed inhibition, however, there was no effect in the ascites tumor cells.

4. Comparative experiments of intravenous injection of No. 23 and No. 26 and intraperitoneal injection of 6MP against subcutaneous sarcoma 180 tumor in mice resulted that No. 26 and No. 23 had more effects than 6MP from the view points of tumor development.

189. THE PRINCIPLE OF THE DAVIS TEST

SATOSHI MATSUMOTO (2nd Dept. of Pharma., Med. School, Kumamoto Univ.)

A new method of the Davis test which is based on the determination of amount of dihydroxyindole, the principle substance, in the urine of patients with cancers was proposed. The clinical result has been shown to be positive more exactly with the cancers, and the consideration is made from the reason why the dihydroxyindole is formed in cancers. In normal body, dihydroxyindole is formed very slightly from indoxyl, and in the cases of cancer, its formation is increased by the decrease of counteraction of indoxyl and the decrease of oxidation of dihydroxyindole in liver. The excretion of dihydroxyindole is also recognized in bacterial infections from the clinical data and also from its formation in principally.

The principle of the Davis test was established from the research of identification of dihydroxyindole, metabolic study on indoxyl and clinical study by improved method.

This work was supported by a grant from the Ministry of Education.

190. POLAROGRAPHIC PROTEIN WAVE OF M_1 -FRACTION OF SERUM IN MALIGNANCIES

NOBUO YAMAGUCHI, TOKIO SASAI, GYOICHI WAKISAKA,
and MAMORU KAKEI*

(1st Division, Dept. of Internal Medicine and 2nd Division,
Dept. of Surgery, Faculty of Med., Kyoto Univ.*)

1) We are interested in the heterogeneity of mucoproteins which are observed in the electrophoresis pattern in the acid buffer (McIlvaine's buffer, pH 4.4, $\mu=0.2$).

2) Using the improved preparative paper strips electrophoresis we prepared M_1 , M_2 , A and B fractions for the polarographic and chemical analysis.

3) In parallel to the increase of M_1 or M_2 or M_1+M_2 , Winzler's fraction of serum also increased, and the sum of M_1 and M_2 covered Winzler's fraction. In pathologic condition (especially liver cirrhosis, nephrosis, malignancies) M_1 and M_2 fractions as well as Wingler's fraction showed the interesting change.

4) The polarographic protein wave of M_2 , A and B fractions and their S.S.A. filtrate waves did not present any polarographic characteristics between cancer and control. But the crossing point of M_1 fraction of cancer serum was higher than normal one. Then M_1 -mucoprotein of cancer serum had the higher 1st wave than the second wave compared to normal M_1 mucoprotein.

5) When Oxyethylcellulose was experimentally added to cystein in cobaltous buffer, the formation and the increase of the 1st wave prior to cystein wave was observed in accord with the rise of oxyethylcellulose-cystein ratio. This pointed to the participation of polysaccharide to the prominent first wave of cancer M_1 -mucoprotein.

6) By chemical analysis M_1 fraction in general, had higher polysaccharide than M_2 fraction. However, there was no significant difference between cancer and normal M_1 mucoprotein as concerned as polysaccharide content.

7) Nevertheless, in regard to the polarographic characteristic of cancer M_1 -mucoprotein, the molecular change of M_1 -mucoprotein including polysaccharide moiety could not be denied.

8) From the polarographic study (native, SSA-filtration, KOH-denaturation) of the experimental mixture of M_1 and M_2 fractions, we could demonstrate that polarographic filtrate wave or Brdicka's filtrate test of serum was composed mainly from mucoproteins of M_1 and M_2 fractions and, in small quantity, from A-fraction.

9) On the basis of the polarographic characteristic of cancer M_1 mucoprotein, M_1 -P.P. test (M_1 -paper electrophoresis-polarography test) was proposed for the detection of cancer.

10) 104 of 186 cancer cases showed positive M_1 -P.P.-test (76.3%): compared to 52.9% of positive polarographic filtrate test. In the non-cancer group only 7.5% of false positivity was showed. The rise of the detection rate in cancer group is explained by the improved detection rate in cases having normal or decreased value of polarographic filtrate test.

191. CLINICAL STUDY ON SERUM MUCOPROTEIN IN LEUKEMIC PATIENT

KATSUHIKO KUBO and GYOICHI WAKISAKA

(1st division, Internal Medicine, Med. Faculty, Kyoto Univ.)

High serum mucoprotein value (Mp) in leukemic patients was already reported by many investigators. In order to observe the interrelation between serum Mp value and clinical feature of the patients, and to detect the qualitative change of serum Mp in leukemia, this study was attempted.

Material were selected hospitalized patients, consisting of 15 acute, 7 chronic myelogenous leukemia, 5 acute, 2 chronic lymphocytic leukemia, 3 monocytic leukemia. Sixty-six measurements were performed on these patients.

The results were as follows:

Increasing of serum Mp in leukemia was observed, and in febrile state, this tendency was remarkable. There were no definite correlation between serum Mp value and peripheral leukocyte count or abnormal cell appearance rate.

Steroid hormone administration was effective to normalization of abnormal serum Mp value and the effect appeared prior to the other symptom, in contrast with that resulted from 6-Mercaptopurine therapy.

Using polarographic filtrate reaction, qualitative change of serum Mp in leukemia patient was reported by Sasai & Kubo.

So, in this experiment, carbohydrate components of pooled lyophilized serum Mp of four leukemic types, that is, acute & chronic myelogenous or lymphocytic, were examined.

Carbohydrate components were examined by Weimer & Moshin (seromuroid & hexose), Rimington (hexosamine), Winzler (sialic acid), and Dische & Shettles' method (fucose), respectively.

Seromucoid and each component of serum protein- and Mp-bound-carbohydrates were increased.

Increasing of seromucoid and protein-bound-hexose and -sialic acid was more marked in lymphocytic leukemia than in myelogenous one, but in both types, high value of each Mp-bound-carbohydrate was obtained and acute form was predominant than that of chronic form.

All component per definite dry weight of leukemic serum Mp were slightly elevated with an exception of hexosamine.

192. COLLECTIVE EXAMINATION FOR CANCER

MASAKA MATSUBARA, TOSHITAKA OHSHITA, FUMIO MITSUI
and MITSUYOSHI HARADA

(1st division of internal Med., School of Med. Toho Univ.)

1) Matsubara reaction is simple in procedure (one physician can examine 500-800 persons per day) and the cost is comparatively low but the biggest feature of this reaction is that the positive rate is high in the early stages of cancer. Therefore, it was thought that the reaction would be useful in mass examination of cancer and the reaction was used, since November, 1958, for examination of a total of 3790 persons of both sexes (including 2067 persons under the age of 30, 1145 persons between 30 and 40, 263 persons between 50 and 60, and 315 persons above the age of 70), who had no subjective symptoms.

According to the result of this examination, 21 out of 3790 persons showed positive reaction and of these, one was found to have chronic myeloid leucosis and six were found to have gastric cancer. Observations are being continued with the remaining 14.

2) Mass examination was also carried out on 595 persons (including healthy and diseased) interned in the Yokufuen (an old people's home) in May 1958. Among the persons examined, 97 have died since then. Comparative examinations were made on the autopsy observations and the result of Matsubara reaction. The reaction was positive in 12, quasi-positive in 8, and negative in 77 out of 97 deaths. Cancer was proved in 9 out of 12 positive cases and 3 out of 77 negative cases. Excluding the quasi-positive cases, this is a positive rate of 91.0%.

Among these 9 positive cases, 5 were healthy at the time of the examination, one had pulmonary tuberculosis, and doubt of cancer had been considered for the remaining two. Out of these 8 cases, three were diagnosed as cancer from autopsy observations and histological examination.

The foregoing results indicate that the Matsubara reaction would be of value as a means for collective examination of cancer.

193. MASS SURVEY OF UPPER G.I. TRACT WITH SPECIAL CONSIDERATION ON THE EARLY DIAGNOSIS OF GASTRIC CANCER

HIDEO IRIE, KÔICHI MURAKAMI, HIROAKI SOEDA and
KEIICHIRO HARADA (Department of Radiology, Kyushu Univ.)

Since 1952, experimental and clinical studies of photofluorographic examination of upper G.I. tract were performed by Irie and co-workers. During 1959 mass survey of upper G.I. tract of the 1416 residents of Itoshima county where the incidence of gastric cancer was the highest in Fukuoka prefecture was done using the method described in this paper.

On the other hand, mass survey of upper G.I. tract of 962 employees of Kyushu University using photofluorographic method was performed in 1960.

In Itoshima county, method of examination and results were as follows:

Among the residents of this area, 1416 persons, who were more than 40 years of age were selected. In this selected 151 persons who had any one of i) positive occult blood in stool, ii) gastric subjective symptom or iii) familial disposition of cancer underwent x-ray examination. 117 of them were taken P.A. and right oblique 28×24 cm² G.I. x-ray pictures without fluoroscopy, 37 of these 117 persons showed suspectable finding. These 37 persons and remaining 34 persons who did not receive x-ray examination without fluoroscopy received fluoroscopic examination. Five gastric cancer, 1 esophageal cancer, 5 gastric ulcers were the major gastric diseases found.

883 employees of Kyushu University who were more than 40 years of age, and 79 volunteers who were less than 39 years old were each taken five photofluorographic pictures. 146 persons showed suspectable findings and 141 received fluoroscopic examination. 51 of them showed positive findings. One gastric polyp, 10 gastric ulcers, 5 duodenal ulcers were the major diseases found.

In Itoshima group 5 gastric cancer were found and none in Kyushu University group. This fact may be due to the difference in incidence of cancer in two groups. Itoshima group contains more older persons than Kyushu University group, and Itoshima county is known to have the highest gastric cancer incidence in Fukuoka prefecture. Kyushu University group is receiving more frequent physical examina-

tion in general, this may be the reason for the difference of number of gastric cancer patients found in two groups.

附 議

友田正信：外科医として胃の集団検診に興味を有するものでありますが、ただいま演者の教室で行われた検診で、胃癌が疑わしいというので、われわれの所に送られた患者で、胃粘膜癌を手術で見出した経験を紹介し、無症状の早期胃癌の発見に果す胃集団検診の意義の大なることを述べた。

194. ON THE OPERATED CASES OF GASTRIC CANCER BY GASTRIC MASS SCREENING

KAIZO ARIGA, KIYOSHI TAKAHASHI, SHOHEI TANAKA and
HIROSHI SATO (Dept. of Internal Medicine School of Med., Nihon Univ.)

57 cases of gastric cancer were found in 10,216 subjects by gastric photofluorography. 28 cases of these cancer were operated and 5 of them were exploratory laparotomy. Gases of SHIMOINA were followed up, and 3 year survival rate was 70%. 4 cases reported were early gastric cancers.

195. STUDY ON THE ACETON SUPERNATANT FRACTION OF CANCEROUS GASTRIC JUICE

KATSUHIKO KUBO, TOKIO SASAI, GYOICHI WAKISAKA
and YASUSHI TAKAGI

(1st division, Internal Medicine, Med. Faculty, Kyoto Univ.)

In 1956, high polarographic activity was detected in Glass' aceton supernatant fraction of cancerous gastric juice by Sasai *et al.*

Since that time, many studies of this fraction were performed in our laboratory. In this paper, nitrogen content of this fraction was measured, by tyrosine method in 139 in- and outpatients including 32 patients of gastric cancer. Gastric juice was collected after histamine stimulation.

The results were as follows;

High tyrosine value was obtained, in general, in acidic juice rather than in anacidic.

And in gastric cancer, the tyrosine value was found, in general, to be high, irrespective of gastric acidity, showing a good agreement with polarographic results in gastric cancer in view of the diagnostic significance.

In order to investigate causative mechanism of high tyrosine value of acetone supernatant fraction obtained from cancerous anacidic juice, proteolytic activity of the anacidic juice was studied at pH 3.5.

Since it was observed that RISA was considerably destroyed by incubating RISA mixed together with the gastric juice, a possible presence of proteolytic activity was confirmed also in cancerous anacid gastric juice.

For the purpose of studying the differences between the normals and gastric cancer patients, if any, ion resin exchange chromatography was carried out, using IR 130.

As a result, some substantial difference could be found between them.

196. STUDIES ON THE NINHYDRIN TEST OF FRACTIONATED GASTRIC JUICE

KENSHI IWASAKI, SETSUAKI KOBAYASHI and MASANOBU AKAGI

(Dept. of 2nd Surg., School of Med. Kumamoto Univ.)

We carried out the Dr. Tazaki's Ninhydrin Test of fractionated gastric juice in 100 cases of gastric cancer and 203 none cancer cases. At the same time, one dimensional ascending paper partition chromatography of fresh gastric juices and their methanol sediments were studied in 18 cases of gastric cancer and 35 none cancer cases preoperatively and same studies were done in 5 cases of gastric cancer and 15 cases of gastric ulcer postoperatively, two dimensional ascending paper partition chromatography of fresh gastric juices and their methanol sediments were studied also in 12 cases of gastric cancer and 6 cases of gastric ulcer preoperatively and same studies were done in 4 cases of gastric ulcer postoperatively.

The results were as follows.

1. In the preoperative groups, 84% of gastric cancers and 45.8% of others were positive in above test.
2. In the postoperative groups, 94% of gastric cancers and 80% of others were positive.
3. It was found that positive agents of this test were chiefly miscellaneous amino acids in gastric juices.
4. The test were always negative with present of free hydrochloric acid even in the cases of gastric cancer.
5. For these reasons, ninydrin test can not be the specific test for gastric cancer.

197. THE CYTOLOGIC DIAGNOSIS OF CANCER USING GASTRIC JUICE

AKIRA ATSUMI and SHYOGO MITO

(Yamagata Prefectural Institute of Health ; Yamagata Prefectural Hospital)

We have tried the cytologic diagnosis of stomach cancer using chiefly gastric juices derived periodically by Katch-Kalk's method.

Examining by acetoanilin staining doubtful particles in 3 testtubes, forejuice, 1 hour juice and 2 hour juice after injection of caffein-solution, the positive rate was 84.5% in 71 cases, which were ascertained histologically as carcinomas. This time we tried two methods of sediment examination, the filtration method and the multiple layer centrifugation method. In the former, the mixture of 3 materials, diluted by physiological salt solution was filtrated by gauze and the filtrated juice removed in glass-filter (No. 4) was absorbed by water stream pump. The material left in filter was centrifugated for 5 minutes by 1500 r.p.m. and then the sediment was examined.

In the latter, the same material was piled on the two Arabian gum layers, each having the specific gravity of 1.030 and 1.065, in the syringe and centrifugated for 5 minutes by 1500 r.p.m.. By this management, cancer cells were revealed to gather in the boundary region between 1.030 and 1.065 and moreover mucus, bacteria and relatively heavy foreign bodies in gastric juice were excluded fairly well.

Our positive rate using two methods mentioned above, each having its merit and defect respectively, was 87.8% in 74 cases of stomach cancer.

Besides, when diagnosis by these methods was yet uncertain, we have selectively used so called abrasive balloon method. Our positive rate in 65 cases indicated 90.8%.

Lastly, roentgen diagnosis, cytologic diagnosis using gastric juice and cytologic diagnosis by abrasive balloon method have their own good points and weak points respectively, so that they help each other in clinical diagnosis of stomach cancer, resulting in high positive rate of 99% in 99 operated stomach cancers.

(千代田生命助成金による)

198. THE TRIALS IN THE CYTOLOGICAL DIAGNOSIS OF GASTRIC CANCER

YOSHIYUKI YOSHIDA and RYO INOUE
(2nd Surgical Division, Med. School, Kyoto Univ.)

Authors' "abrasive balloon with a sheath" (modified Panico's abrasive balloon) was presented as an improved instrument of obtaining fresh, well preserved and relatively selective gastric specimens. The apparatus consists essentially of single-lumen polyvinyl tube which ends in two pieces of net-covered inflatable balloon, and a sheath which sheathes the tube. The balloons are kept in the distal thicker portion of the sheath while the tube is passing through the oesophageal tract. The polyvinyl tube is designed to be just firm enough not to injure the stomach mucosa or not to be curled in the stomach but to be pushed down to the pylorus under control of examiner's hand. The sheath should have also the flexibility not to injure the surface structure of the oesophageal tract.

When the instrument is used, it is first inserted in sitting position, and once the tip of the sheath reaches the lower end of the esophagus, the proximal portion of the instrument is held with examiner's hand, and the polyvinyl tube, to which two balloons are attached, is pushed out gently. Then the mucosal surface of the stomach is abraded. After the abrasion the balloon is drawn into the sheath, and then the whole tube is removed from the patient. The balloon reaches the pylorus without the aid of peristalsis. This procedure can be accomplished without the control of the fluoroscopy.

The gastric cytology of the 139 patients was studied by means of three kinds of abrasive balloon techniques (i.e. the first group with Panico's abrasive balloon, the second group with the authors' "abrasive balloon without sheath" and the third group with the authors' "abrasive balloon with a sheath"). The results in 119 cases to which diagnosis had been confirmed postoperatively were reported.

The following conclusions with the authors' balloon technique with a sheath: i) The invasion of many cells and substances exfoliated from the oesophageal tract into the specimen was prevented. ii) The loss of the specimen obtained from the stomach was prevented. iii) Abrasive balloon was able to reach the tumor surface without contamination with detached cells and substances from the oesophageal tract. iv) It was easily accessible to antral and pyloric regions without the aid of peristalsis. v) The authors' apparatus is simple and safe, and certifies less time consuming in cytologic examination.

For more precise the identification of malignant cells, the specimens obtained from

the stomach are stained by several histochemical staining methods. In these studies, it is shown that acid phosphatase staining is available for the identification of suspicious cells. The microphotographs of gastric specimen stained by acid phosphatase technique were presented.

199. A MASS EXAMINATION FOR DETECTION OF RECTAL CANCER IN AOMORI PREFECTURE

MITSUE MIURA, KEIICHI ONO, TOMONOBU SATO, HIRONOBU NAKAYAMA,
YUKIO TAKEDA, MASAOKI KIMURA, MITSURU TOGASHI, MITSURU KON,
CHUZO OHTA, SUSUMU SHIBATA, YUKINAO HARIMA
and YASUYUKI MARUMI

(Surgical Clinic, Faculty of Med., Hirosaki Univ.)

Mass examinations were carried out at 15 points in Aomori Prefecture to detect rectal cancer from November, 1959, to May, 1960. 1,003 subjects were examined, of which 422 were male, and 581 were female. About 60 per cent of them were over 30 years of age. The majority of the subjects examined had complaints such as uncomfortable feeling in the lower abdomen, anal pain, obstipation, rectal or anal bleeding, etc.. Digital and anoscopic examinations were performed, and in some cases also romanoscopy and biopsy.

Five cases (0.5%) were diagnosed as rectal cancer. They were all females over 40 years old, and 2 cases of them were histologically primary rectal adenocarcinoma, and 3 cases were infiltrated cancer which had developed from uterus cancer. Twenty-four other cases (2.4%) were diagnosed as rectal polyps by anoscopy, 8 cases of which were confirmed by the extirpation or biopsy. Biopsy was performed also in one case suspected clinically of carcinoma, but it was found histologically to be a chronic inflammatory tumor.

From the above results, which showed a higher percentage of rectal cancer than pulmonary cancer, the authors believe that mass examination is necessary for the early detection of rectal cancer.

200. CLINICAL ASPECT ON THE GROWTH OF LUNG CANCER

YUZO TAZAKI and HITOSHI TOMINAGA

(Cancer Institute Hospital)

The volume of tumor depends upon the duration of the period of growth and the

rate of growth. If the time interval and change in volume are known, the average growth rate can be determined, the duration of a given tumor and the time of inception can be estimated. According to this hypothesis, introduced by Dr. Collins, a single cancer cell 10μ in diameter will grow to a nodule 1 mm in diameter in 20 doublings. A 1 cm nodule would be achieved in 30 doublings.

12 cases of primary lung cancer which were treated as pulmonary tuberculosis for a certain period were collected to be adequate for the estimation to observe the rate of growth. In some instances, however, their growth was not always hypothetical. Slowly growing tumors were observed in 3 cases and rapidly growing tumors in 3 cases.

In one of rapidly growing cases, doubling time was estimated as one month. In slowly growing tumors, doubling time was too slow to be estimated. In one of other cases, doubling time was calculated 3 months. The single cancer cell of this case can be supposed to be generated 8 years ago.

附 議

宮地 徹：興味の多い御研究ですが、御指摘のような肺癌結節を組織学的に御検討いただいて Collins の理論に合うかどうかみられたでしょうか。私見によれば、人の肺癌はたとえ球形にみえても固有の肺組織を含んでおりその量もまちまちだと思うからです。

肺癌が臨床症状を呈するまでに長年月を要するだろうという御推測には賛成いたします。私どもの経験でも、肺結核治療中に発生した肺癌がその症状を現わすまでに3年を要しておりました。

富永：組織学的に、肺癌組織のなかに癌細胞がどの程度含まれているか検討しておりませんが、癌組織がレ線的に限局して腫瘍状に発育していると思われる症例をとり上げています。また、癌組織内に癌細胞数に一定の割合で間質等が含まれているとすれば、レ線的に経過をみると対象グラフで直線状になります。このときは1コの細胞の大きさが変わったとしてとり扱えます。不規則に間質等が含まれてくるとすればレ線的にも不規則な増大を示すものと思われまゝ。もし、癌細胞の大きさが 10μ で、細胞1コに対し約2割の間質が規則的に含まれると仮定すれば、直径1cmになるまで29回の分裂でよいという計算になり、間質等が全然ないと仮定した場合の同様の分裂回数が30回に較べて、大差を示しません。

201. CLINICAL STATISTICS ON LUNG CANCER

YUZO TAZAKI, ICHISUKE FURUKAWA, SEKIJI MIYAJIMA,
HISASHI FURUE, YASUSHI INADA, HITOSHI TOMINAGA,
JUNTAI SATO, MAKOTO GOMI and FUMIO TAGAYA

(Cancer Institute Hospital)

Clinical statistics has been made on the 263 cases proved to be of primary lung cancer at our hospital for these ten years.

The ratio of male to female is 3.3 to 1. For the age, the largest number is found in the decade 60-69 years, somewhat fewer are in the decade 50-59 years and appreciably fewer (4 to 5%) in the decade 30-39 years. Difference of the age

distribution between male and female is negligibly small.

Heredity: 34% of the 265 cases has cancer patients within a relation of the third degree.

Habit of smoking: more than 90% of the male patients has the habit of smoking, and furthermore, 12% of them smoke more than thirty cigarettes a day.

As for the duration from the first beginning of symptom to the first medical examination, any difference is not seen in the patients of hilar type and those of peripheral type. On the whole, in case of hilar type the duration from the first medical examination to the proved diagnosis seems longer than in the peripheral type.

Such clinical symptoms are found: cough (43.4%), sputum (24.2%), bloody sputum and hemoptysis (18.5%), fever and chest pain.

At the first examination, considerable numbers of the patients complain of swellings in cervical lymph node, and of hoarse voice as the symptom of metastasis. On the other hand it must be noted that there are 11.6% of the patients whose lung cancer is found among the people with no complaints at the mass screening.

According to the comparison between x-ray finding and the symptoms at the first medical examination, cough is observed in 68.5% of the patients, and bloody sputum and sputum follow it, and the patients without complaints are not found among the hilar type patients. On the contrary, no complaints are found in 28.1% of the peripheral type patients.

202. STATISTICAL OBSERVATION OF GROUP EXAMINATION FOR THE UTERINE CANCER, DONE DURING PAST 6 YEARS

AKIRA TABUCHI and ATSUSHI FUJIWARA

(Obst. & Gyne. Dept., Med. School, Hiroshima Univ.)

Our Department has performed group examination for the uterine cancer, which serves as well for an education movement of cancer with cooperation by Health Centers and Local Women Societies in various districts 9 times up to present since 1955.

We are going to report the statistical observation on the items of investigation and the results of the examination with colposcopy, cytology and histology on 2796 women, who have been examined.

- 1) Many subjects ranged between the 4th and 5th decade.
- 2) Predisposition of cancer was seen in 29.2% of the subjects.
- 3) Colposcopy revealed a tendency that cervical erosion recedes with aging, namely, squamocolumnal junction gradually recedes toward cervical canal.
- 4) Abnormalities similar to cancer cells were seen by cytological examination in

many of those, who had senile vaginitis or histologically atypical hyperplasia.

5) Eight cancers of the portio vaginalis were detected by histological examination.

203. ACTIVITY OF THE ORGANIZATION FOR EARLY DIAGNOSIS OF THE UTERINE CANCER

YOSHIO ASHITAKA, SHINZO ISOJIMA, AKIRA YAGI*,

NORIO AMAKI* and TERUKO HASEGAWA*

(Dept. of Gyne., Med. School, Osaka Univ.; Toyonaka Health Bureau*)

The best way of fighting against cancer is to find out the patient in precancerous stage or stage 1, and to treat them without delay. Osaka Prefectural Government set up the diagnosis clinic for the uterine cancer in eight health bureaus in the main cities, and opened them once a week for women. This is a report from the Toyonaka Health Bureau to which about 200,000 people belong.

1. For 15 months, 275 women visited the health bureau for the examination and ten cancer patients (3.6%) were found out. As epithermoid carcinoma of cervix, one stage-0, four stage-I, one stage-II and one stage-III patients were found. As other type, one adenocarcinoma (corpus uteri), one carcinoma of vulva and stump recurrence of carcinoma after the radical operation were noted.

2. The City News and other news paper contributed greatly to make women understand the necessity of examination but doctor's or women's societies did not show any helpful influence over them.

3. A peak of age distribution curve of visitors was at a range between 35-39 years old, and even at peak the number of visitors was less than 1% of female population in the same age.

4. 16.7% of visitors had no symptom but others had some; irregular vaginal bleeding (27.6%), contact bleeding (10.9), discharge (48.1%) and lumbago or lower abdominal pain (22.2%).

5. Only two out of 10 cancer patients had remarkable subjective symptom and five had slightly. Six out of 10 cancer patients had macroscopically some abnormal findings and five out of 9 cancer patients had colposcopically pathologic findings. But all of cancer patients showed cytologically positive results.

附 議

河津竜介: われわれは子宮癌の早期診断, 早期治療を目的として本年5月より熊本県玉名郡菊水町をモデル地区に選び毎週土曜日に現地向って集団検診を行っているが12月3日までの成績を発表した。

検査回数 27 回検査総数 519 名である。この中子宮頸癌 4 名 (0.8%) 子宮腔部癌 213 名 (412.2%) であった。

細川 勉：われわれは昨年本学会でわれわれの集団検診の成績を発表したが、現在までの成績では約 2200 名中 3 名の癌を発見している。この発見率から集団検診により多数の癌患者を発見するということを期待することには限度があり、むしろ定期検診の必要を啓蒙する意味に意識を感じなければならない。都内の一流会社、官庁職員の希望者検診でもその感を深くしている。

204. CLINICAL RESULTS OF COLPOMICROSCOPE REFORMED AND IMPROVED BY THE AUTHORS

HAYAMI FUJIMORI, HIROSHI KINOSHITA and SADAMU NODA

(Dept. of Obst. and Gyne., Med. School, Osaka City Univ.)

It is said that the Colpomicroscopy has many superiorities for the early detection of cancer of the cervix, compared with other methods. Antoine's Colpomicroscope is trouble sometimes to operate. And so a new style of Colpomicroscope was reformed and improved by the authors. Using the Colpomicroscope of this type many effective and excellent results were obtained, comparing with other methods, for instance the cytological, histological examination etc. .

IX. Radiation

205. STUDIES ON INACTIVATION OF FRUCTOSE SARCOMA CELLS BY UV-IRRADIATION

TSUGUO KUWATA, YOSHIHIRO YASUMURA and MASAYOSHI KANISAWA

(Dept. of Bact. & Path., School of Med., Chiba Univ.)

Fructose sarcoma can be trypsinized and easily dispersed into single cells. Accordingly, it is possible to titrate viable tumor cells thus obtained by intracerebral inoculation method as reported before (GANN 50, Suppl.: 132, 1959). Four ml of cell suspension, which contained about 2.0×10^6 to 8×10^6 cells/ml, was put into Petri dish and irradiated with "National" germicidal lamp from 40.5 cm distance. During irradiation, Petri dish was shaken by hand. Then, small samples were taken out and viable cell fractions were measured. Under these conditions, whole cells lost their viability after 40 second irradiation, despite their non-stainability with eosine. However, when normal embryonic tissues of mice were added to tumor cell suspension thus inactivated, then tumors appeared in the subcutaneous space of transplanted mice. In the early stage of tumor development, tumor cell colonies were detected among embryonic tissues. In the later stage, tumors became entirely sarcomatous and parts of embryonic tissues disappeared. Probably, few viable tumor cells, which escaped from UV-inactivation, multiplied under the influence of normal embryonic tissues and made tumor cell colonies. However, other mechanisms should be also considered. By adopting this method, we have obtained a subline of fructose sarcoma which is more resistant to UV-irradiation than the parent line and moreover causes lung metastases in mice. Fructose sarcoma never produced metastases without cortisone. This new subline differs from the parent sarcoma cells in their convertibility to ascites form. Details of ascitic conversion process will be reported elsewhere.

206. STUDIES ON THE DESTRUCTION OF ASCITES TUMOR CELLS BY INTENSE ULTRASOUND

MATSUZO TSUCHIDATE, TSUNEYOSHI NAGASHIMA, TOSHIO WAGAI
and SHUICHI HAYASHI

(Dept. of Surgery, School of Med., Juntendo Univ.)

The destructive effect of intense ultrasound energy to ascites tumor cells were studied using AH130, AH13, AH7974 and Yoshida sarkoma. Ultrasonic radiation in our experiments were carried out *in vitro* and *in vivo* which the whole body of rats are radiated in water tank. In this paper the studies *in vitro* are mainly reported.

The ultrasonic generator used in this experiments is constructed so that we may choose any frequency of 400KC, IMC, 2MC, 4MC, and power of 80, 120, 300, 500W. The ultrasonic transducer is made of barium titanate which is formed rectangle 35×100 millimeter in size and in order to obtain a wide ultrasonic field we used the reflector of ultrasonic wave.

As the results of our experiments, we could observe the difference of ultrasonic destructive effects to the structure of various tumor cells between AH130 and other ascites tumor cells by the simple ultrasonic radiation. And then AH130 were destructed completely within 10 minutes, but the other tumor cells were less destructed compared with AH130 using a frequency of 1 megacycle ultrasound and power of 500 watt.

On the other hand, the application of ultrasonic cutting method to this experiments was excellent successful for the destruction of tumor cells by ultrasonic radiation. We radiated tumor cells together with colloid gold, carbon, quicksilver and so on. In this procedure gold and quicksilver was most effectual for the destruction of all kinds of tumor cells.

Moreover in the series of this studies, human blood cells were not damaged by using similar ultrasonic power and frequency.

207. A CYTOLOGICAL OBSERVATION ON CHROMOSOME BREAKS IN MN LYMPHOSARCOMA CELLS, INDUCED BY X-IRRADIATION

ASAO INUI and TOSHIHIDE TABATA

(Dept. of Otorhinolaryng., Wakayama Med. College)

The present study was attempted to inquire the chromosomal aberration, especially chromosome breakage, induced by X-irradiation in the tumor cells of mice.

For materials the transplantable lymphosarcoma occurred on Na₂ mice were utilized in ascites from.

X-irradiation was given on the tumor-bearing mice under the following conditions: Deep therapy installation 180 KVP; filter of 1.0 mm Cu and of 0.5 mm Al; five different dosages such as 2000r, 1000r, 400r, 200r and 100r.

The tumor stem cells of MN-lymphosarcoma were characterized by 40 rod-shaped chromosomes.

Chromosomes of tumor stem cells were observed by acetic orcein squash technique immediately after irradiation.

Remarkable chromosome breaks were induced by X-irradiation with high doses such as 2000r, 1000r, 400r and 200r, but the cells were unsuitable for cytological analysis under those doses, because the chromosome breaks were too many.

The most suitable X-ray doses for cytological observation was 100r.

Idiograms of 100 tumor cells containing chromosomal breaks were analysed. As a standard for measurement of each chromosome, the largest chromosome length in metaphasic plates of stem cell was used. According to this method chromosomes of stem-cell were statistically classified into seven groups such as 0.4, 0.5,1.0.

These orders represented the chromosome length. Idiogram analysis showed that chromosomes classified into 0.9 orders had the highest frequencies of breaks than others, that the location of breaks are represented by a calculated value of f/a (a =length from Kinetochore to tip of the chromosome; f =length from Kinetochore to location of breaks).

Most of the chromosome breaks were adjacent to the middle of the arms. It may be concluded that, among the stem cell chromosomes of MN-lymphosarcoma, those having the ordered length of 0.9 are most sensitive to X-irradiation, and that the radiosensitive region is close to the middle portion of the arm.

208. THE CYTOLOGICAL STUDIES OF MN-SARCOMA (II) THE MITOTIC INHIBITORY EFFECT OF X-IRRADIATION

MAKOTO MAEDA and ICHIRO NISHIBATA

(Dept. of Otorhinolaryng. Wakayama Med. College)

The authors investigated the mitotic inhibitory effect of X-ray single irradiation on MN-sarcoma cells of Na₂-mice in ascites form at the point of mitotic index. The X-ray used in this investigation were conditioned as follows; secondary voltage 180 KVP, secondary current 15 mA, filtration with Cu 1.0mm+Al 0.5mm, focal distance 40 cm and 43.9 r./min. The dosis of 200 r., 300 r. and 400 r. of X-ray were irradiated on the tumor bearing animals on the 3rd day after inoculation. The effect of X-irradiation on mitotic rate fell into 2 stages; (a) stepwise decline of mitotic rate, (b) prolonged mitotic recovery. In 400 r. irradiated group, a period of complete mitotic inactivity was showed at 2 hour after irradiation.

The followings were considered from the change of the each proportion of the mitotic phasis in 400 r. irradiated group: (a) No cells newly entered the mitotic process after irradiation. (b) The cells which were in the mitosis would go on the dividing process and reach to complection. (c) The early prophasic cells at the irradiation would reverse the mitotic process and return to the resting stage.

Approximately 3 hour after irradiation, mitosis reappeared in the ascites, and showed gradual increase. And 8 hour after irradiation, mitotic indices recovered to the nearly preradiative condition.

209. EXPERIMENTAL STUDIES ON ACQUIRED RADIO-RESIS- TANCE OF TUMOR CELLS (SEQUEL REPORT)

SHOICHI OBOSHI and TATSUYO SHINOZAKI

(Dept. of Path., of Radio., School of Med., Hirosaki Univ.)

At the annual meeting of the Japanese Cancer Association in 1959, the authors reported that the tumor cells, which were consecutively treated with the irradiation of relatively smaller doses of X-ray for a long period, showed no development of radio-resistance. In the present study, further investigations were performed as to whether the tumor cells treated with the intermittent irradiation of relatively larger doses of X-ray acquired a radio-resistance or not.

The original diploid line of Hirosaki sarcoma, an ascitic lymphosarcoma of rat,

was used in common with the previously reported experiment. When the ascitic tumor cells increased in number, a whole body irradiation of the doses of 1,000 r was carried out and then the tumor cells were transferred. Such procedures have been undergone over 34 generations during 190 days. The total doses of X-ray hitherto irradiated amounted to 34,000 r. The cellular effects of 200 r and 500 r of the X-ray irradiation, respectively, were employed as a criterion of a development of radioresistance of the tumor cells. The fall of the mitotic rate of the irradiated variants, which were derived at 75 th, 140 th and 190 th day after the beginning of the experiment, respectively, was completely identical to that of the original line.

From these results including the previously recorded experiments, it was concluded that the tumor cells, which were consecutively or intermittently treated with the X-ray irradiation for a long period, showed no development of radio-resistance. Therefore, the acquired radio-resistance clinically observed may be due to the altered environment in which the tumor cells are settled. (文部省科学研究費による)

210. EFFECTS OF X-IRRADIATION UPON ENZYME SYSTEMS OF TUMORS

KEIJI HORI, SUTEHARU MOCHIZUKI and YASUSADA FUJINO

(Dept. of Radio., Med. School, Osaka Univ.)

Alterations in the functions of enzyme systems of TCA cycle, oxidative phosphorylation, phosphatase groups, nucleic acids etc. of experimental tumors after X-irradiation were recently reported by authors and other investigators. This time we have examined these enzymic changes *in vitro* in Warburg's apparatus, 37°C, irradiating of ^{60}Co - γ rays (about 1000r/min, and on the other hand electro microscope studies were carried out.

Results are as follows.

In vitro after 30 minutes to one or one and half hours, enzyme activities of succinic dehydrogenase, RNAase, DNAase show no appreciable changes. TCA cycle's enzyme system seems to decrease 10 to 15% 30 minutes after irradiation, and seem to be more raised one to one and half hours. The value of oxidative phosphorylation also decreases about 15% after one hour, but the decrease seems to appear later than the former. ATPase increases after one to one and half hours about 20%, but 5-nucleotidase are not appreciably increased.

Incorporation of ^{32}P into RNA of microsome fractions seems to increase one to 2 hours after irradiation.

Morphological changes of mitochondria are found electron-microscopically from 30 minutes and swelling, vacuoles and unidentified bodies in mitochondria are seen one to 2 hours.

These morphological changes seem to appear more severely after total body X-irradiation and the effects seem to be stronger by indirect elements. But these histological changes seem to clarify the chemical results of TCA cycle, oxidative phosphorylation etc..

附 議

前田 真：核分裂像より見れば 10 分後には変化がきている。あなたの実験は 30 分 1 時間といわれたが、もっと早く見る必要がある。(弱いレ線量では、5 時間後には完全に、レ線影響から解放されることも知る必要がある)。

堀：ミトコンドリアの場合、電顕的には染色体の時のように 10~15 分では変化が認め難いように思われますが、さらに検討してみたいと考えます。

211. BIOLOGICAL EFFECTS OF SO-CALLED IRRADIATION TOXIC SUBSTANCE (C-X SUBSTANCE)

SHIGEKAZU MAEDA (Dept. of Obst. & Gynec., Osaka City Univ.)

The aqueous layer obtained by the removal of protein from the serum of irradiated rat or rabbit by centrifuging, adding 0.3 N Ba(OH)₂ and 5% ZnSO₄, was developed on the paperchromatograph. From one spot (Pf: 0.69) an unknown substance named C-X substance was obtained. This substance showed positive reaction on biuret, ninhydrin reaction, and negative on xanthoprotein, Hopkins-Cole and Millon reaction on the contrary.

Biological characteristics of C-X substance

- 1) 1 mg of C-X substance was injected everyday into the peritoneal cavity of mice, and decrease in liver catalase activity was recognized on the 8th day by measurement with KMnO₄.
- 2) The uptake (c.p.m.) of ⁵⁹Fe in blood, liver and kidney of mice treated with C-X substances was higher than that of the control mice.
- 3) No difference between treated mice with C-X substance and non-treated one was recognized by bromsulfarein test.
- 4) The value of glutamic oxaloacetic transaminase and glutamic pyruvic transaminase of rats treated with C-X substance intraperitoneally was higher than the control.
- 5) No histological change was recognized between treated mice with C-X substance and non-treated ones.

212. CLINICAL STUDIES OF "OX SUBSTANCE" (I)

DENNOSUKE JINNAI, SANAE TANAKA, JUNYA SHIMIZU,
MASAKAZU ONO, KUNIO OKAJIMA, JUN'ICHI KOBAYASHI
and YOSHITOMO KUWAHARA

(Dept. of surgery, Med., School Okayama Univ.)
(The Cancer Institute of Med. School, Okayama Univ.)

"OX substance" is a fraction of unsaturated fatty acids extracted from the liver of X-ray irradiated rabbit. After many biological tests concerning the properties of "OX" by Yamamoto *et al.*, it has been found that this substance possesses an anti-carcinogenic power as well as an inhibitory property on sperm formation but has no damaging effect on bone marrow and parenchymatous organs. With the purpose to determine the efficacy of OX it has been tried in the treatments of 148 cancer patients. As OX is insoluble in water, we use it in the form of OX-Na compound regulating its pH at 7.2 for local application, but for the intravenous administration in the form of OX colloid solution.

The local application of this substance dramatically eliminates carcinomatous stenosis in esophagus, cardia, rectum, and urinary bladder. In the case of bronchial carcinoma likewise the introduction of this substance through trachea improves the general and local findings both roentgenographically and clinically. And fluid in pleuritis and peritonitis carcinomatosa is markedly eliminated by local administration. Next, the intra-tumoral injection of OX brings about atrophy or abscess formation of tumor in plane surface carcinoma. Thus in 75 cases alleviation due to this local application can be seen in 92 per cent. In 103 cases given intravenous administration atrophy and inhibition of the proliferation of tumor can be observed. In the cases of gastric carcinoma and others, carcinoma tissue shows diffuse necrosis and tumor itself is flattened or undermined. Histologically there are polynuclear giant cells in tumor tissue. In the case of squamous cell carcinoma there can be recognized calcification of carcinoma cell nests and in others the atrophy of tumor can generally be observed, yielding objective good results in 33 per cent of the whole. From these findings it is assumed that a more effective result can be expected with a further modification of OX itself.

附 議

前田茂和: OX 物質の抽出法いかが。

入江英雄: OX 物質が大変よいというのに学界一般の追試も少く一般に認められないのは何故か。

演者: ① 抽出方法についての答

OX は不飽和脂肪酸の一種で、健康家兔に X 線を照射した後、肝ホモジネートから一般の不飽和脂肪酸の抽出法にしたがって分画することができる。

③ 座長からの質問についての答

OX は局所的には著明な選択的制癌効果を有することを示したが、静脈内投与が行われない限り制癌剤としての意義も少い。不飽和脂肪酸自体には多少の溶血性現象があり、かつこのような油状物質をコロイド化して静注投与を行うためには、かなりの研究を要し、現在改良を重ねつつあるが、安全なものができるれば、いづれ御追試をお願いすることになるだろう。

陣内：私どもは各種の制癌剤の使用の経験をもっていますが、この OX 物質は他の制癌剤に比し顕著な効果をもっており、組織学的にみましても癌組織のみに選択的に作用するというこの他、巨細胞の出現が非常に著明であります。他の制癌剤でも大量やれば巨細胞も出現しますが、この OX ではわずかな量でも常に巨細胞の出現をみるということでもあります。

また骨髓機能の抑制がありませぬために、他の制癌剤で白血球減少がきましたものにつづいて OX を使用しましても、ぐんぐんと白血球数も増してまいります。

この一月前にできました静注用のものは副作用がほとんどなくなりましたのでやがて皆様のお手許にお届けすることができると存じますので、何卒御追試いただいて御忌憚なき御批判を仰ぎたく存じます。

213. X-RAY THERAPY OF CARCINOMA OF THE PARANASAL SINUSES AND NASAL CAVITIES

KAZUO HARA, YASUSHI HAMASAKI and SHUN'ICHI SAKAI*

(Dept. of Radio., Dept. of Otorhinolaryng*, Med. School Osaka Univ.)

We have investigated 123 primary cases of carcinoma of the paranasal sinuses and the nasal cavities, which we have been concerned with for recent 4 years. Our radiological examinations of these cases were consisted of taking routine films and tomograms. According to the grade of the tumorous invasion we have fixed the therapeutic plan. Soon after exploratory excision had been done, the X-ray irradiation (200 kvp) was applied to rather wide area (10×7-8cm), extended from upper orbital edge to lips, with 4 fields, each of which was given 250-300 r/day and 2000 r (air dose) in total, within 4-5 weeks. About one and half a months after radical operation and intracavitary radium therapy were carried out.

The X-ray irradiation alone has been applied to inoperable cases, which have signs as follows:

- 1) cases showing the infiltration to the cranial basis.
- 2) cases showing the destruction of the pterygoid process.
- 3) cases showing the destruction of the temporal bone.
- 4) cases showing considerably bilateral spread.
- 5) cases showing the wide destruction of the anterior wall of the antrum. In these cases each field was given 2200-2500 r or more.

One of the authors, Y. Hamasaki, has roentgenologically divided carcinomata in this area into 6 groups, thereafter considering of depth and width of the infiltration divided each in to 2 types.

Results

("X-ray irradiation alone" contains operable cases, those rejected operation.)

1-year crude survivals	irradiation alone	29 in 40 cases	(70 %)
	operation combined	14 in 25	(56 %)
	total	43 in 65	(65.4%)
	Type I (early stage)	18 in 23	(78.3%)
	Type II (late stage)	25 in 42	(59.5%)
2-year crude survivals	irradiation alone	8 in 20	(40 %)
	operation combined	9 in 20	(45 %)
	total	17 in 40	(42.5%)
	Type I	11 in 15	(73.3%)
	Type II	6 in 25	(24.0%)
3-year crude survivals	irradiation alone	3 in 10	(30 %)
	operation combined	6 in 14	(43 %)
	total	9 in 24	(37.5%)
	Type I	7 in 10	(70 %)
	Type II	2 in 14	(14.3%)

Trying to find out the carcinoma at the earlier stage, we can probably level up the survival rate by use of the X-ray irradiation alone.

附 議

前田：レ線治療後に手術をする場合、その間隔を6週間とするのは、長すぎると思う。癌細胞の再増殖の上からは早ければ、早い方がよいのである。

原：1) 術前の治療線量が多いために、皮膚反応の回復をまつのに1カ月半の期間をおいています。原則としてもつと早い方がよいという御意見に対しては賛成ですが、照射量が多いために1カ月半の期間は制癌の状態にあると考える。

山下久雄：1) Radium 療法は何の位の線量を与えられましたか。なるべく大量を早く照射するのがよいと考えています。

2) われわれのところでも、同じような術式で治療していて、5年治癒率約40%を得ています。いろいろの問題点があると考えられますが、術前照射線量と照射手術間隔との関係が一番大切だと思います。われわれはCo-60大量照射を利用して皮膚障害を少くし、手術による皮膚の損傷を少くして、もっと早く手術の方がよいと信じます。もっとも、1.5カ月の間隔はそんなに長いとは思わない。十分な照射をすれば、それでも差支えない。

原：ラジウム治療は術後2000 mghr 最近では3000 mghr 投与しています。

214. RESULTS OF RADIOTHERAPY OF CANCER OF THE TONGUE

HISAO YAMASHITA, SABURO AMINO and MAKOTO GOMI

(Dept. of Radio., Cancer Institute Hospital)

It has been considered that the results are best for tumors of the tongue, in case

when the radiation therapy is applied, owing to the suitability of the location. We have examined the clinical conditions, assumed them as important factors to exert the results.

During September, 1946 to March, 1955, a total of 190 new cases, diagnosed histologically as malignant tumor, have been treated radiologically, without any other previous treatment, at the hospital of Cancer Institute. Up to March, 1960. all cases have already been over 5 years.

Classification: The patients were staged, based on the I.R. C. Grade, accepted in 1953, when admitted to, or when first seen, at the hospital as follows: Stage I-22 cases, Stage II-49 cases, Stage III-22 cases and Stage IV-97 cases.

Therapeutic method: Intratumoral insertion of radium needles was principally used for most cases of the cancer of the tongue and in some cases, radon-seeds implantation, external irradiation or their combined therapy was applied. Neck dissection was performed for regional lymph node metastases.

Results: The survival rates after 5 years, classified in grades, of these cases were 86.3% (Stage I), 75.5% (Stage II), 50% (Stage III) and 14.4% (Stage IV). The survival rate as a whole was 42.6%.

215. RADIATION TREATMENT OF CANCER OF THE ESOPHAGUS

HISAO YAMASHITA and HIDEO KOBAYASHI

(Dept. of Radiol., Cancer Institute Hospital)

Four hundred and twenty-eight cases of cancer of the esophagus had been treated at the Japan Cancer Institute Hospital since the reopening of the hospital after the war up to November 1959. Subdividing these cases into three groups, upper one third (cervical region), middle one third (neighbourhood of bifurcation), and lower one third (hiatus region); and further to sex and age, the results are given in Table 1. In the cervical cases females are higher than males, but in the two other cases males markedly overnumber the females. And in the age rank the fifties are at the top in upper and middle cases, but in lower cases the sixties are remarkably numerous over other ages. In the whole the sixties are the highest in number with the fifties coming up next.

Histopathological examinations had been confirmed in 232 cases. The majority of cases (223 cases=96%) were squamous cell carcinoma. The other 8 cases were found to be adenocarcinoma and one case reticulum cell sarcoma.

As methods of irradiation depth x-ray irradiation and Co-60 teletherapy were chiefly taken to. With x-ray irradiation crossfire radiation from 3 fields was mainly

applied to cervical cases, and to the others rotating radiation was usually given. With Co-60 teletherapy, 2 fields cross-fire radiation or rotating method were took to in lower cases.

Irradiation dose: At the earlier periods a 3000 r mark was taken, but more recently an increased 5000 r mark is being observed.

Radium treatment and radon-seeds implantation are at present applied only as supplementary means; both of which had long been applied as main therapy in former days, and hardly been obtained good results.

The rate of occurrence of metastasis in lymph nodes is highest in the supra-clavicular region, the discovery of which is easily made. Among the total 42 regional metastasis 35 cases belong to cervical ones, that is 40% of the whole cervical cases; 6 cases to the middle one third (2.5%), and 1 case to the lower one third (1%). In Table 2 the occurrences of metastasis and the complication are shown briefly, which heavily depend upon the prognosis of the patient.

The results of the treatment are summed up in Table 3. The primary cure rate and one year survival rates are relatively high, but the survival rates in the third and further years are remarkably declined. While the radiation therapy of esophageal cancer can be said to be effective in most cases, the true cure are only achieved in few cases. The study to determine the adequate amount of the tumor dose and its time-dose relationship, and further the effective treatment for metastasis and complications, still remain for us to tackle.

Table 1. Sex ratio and age rank in cases of cancer of the esophagus

Location	Sex	Age rank					Total
		Thirties	Forties	Fifties	Sixties	Seventies	
Cervical regions	Male	4	3	16	15	4	42
	Female	4	14	11	11	7	47
	Total	8	17	27	26	11	89
Middle one third	Male	2	17	62	109	29	219
	Female	0	2	10	14	9	35
	Total	2	19	72	123	38	254
Lower one third	Male	0	6	26	27	15	74
	Female	1	1	5	1	3	11
	Total	1	7	31	28	18	85
Total	Male	6	26	104	151	48	335
	Female	5	17	26	26	19	93
	Total	11	43	130	178	67	428

Table 2. Metastasis and Complications of cancer of the esophagus

Location	Lymphogenic metastasis		Haematogenic metastasis	Complications	
	in regional glands	in distant glands		Bleeding	Perforation
Upper one third	35 (40%)	—	4 (4.5%)	4 (2.5%)	1 (1%)
Middle one third	6 (2.5%)	35 (14%)	16 (6.5%)	6 (2.5%)	15 (6%)
Lower one third	1 (1%)	9 (11%)	2 (2%)	—	1 (1%)

Table 3. Results of radiation therapy of cancer of the esophagus

Location	Therapy methods	Primary cure	Survived				
			1 year	2 years	3 years	4 years	5 years
Upper	5,000 r and more	57 : 16 (28%)	57 : 16 (28%)	50 : 7 (18%)	46 : 5 (14%)	37 : 2 (5.5%)	26 : 1 (4%)
	Less than 5,000 r	32 : 2 (6%)	32 : 3 (9%)	23 : 0 (0)	20 : 0 (0)	18 : 0 (0)	17 : 0 (0)
Middle	X-rays cross-fire	95 : 31 (33%)	95 : 10 (11%)	95 : 4 (4%)	95 : 3 (3%)	95 : 3 (3%)	89 : 3 (3.5%)
	X-rays rotation 4,000 r and more	111 : 11 (10%)	111 : 9 (9%)	76 : 0 (0)	58 : 0 (0)	18 : 0 (0)	—
Lower one third	Co-60 teletherapy	65 : 11 (19%)	65 : 11 (19%)	61 : 3 (5%)	52 : 2 (4%)	39 : 1 (2.6%)	24 : 0 (0)

附 議

梶林和之：転移果の存在を予想して、その部に予防的放射を併用していますか。

野口忠之：1. 適切な深部線量は何 r 位なりや。

2. 適切な深部線量は固定照射にても回転照射にても同じですか。

3. 肺線維症の出現率ほどの程度か。

小林：① 梶林先生え

転移の治療は放射線または手術によって行っているが、転移想定部位に対する予防照射は行っていない。

②

12000~14000 r というのは空中線量でこれを病巣線量に換算しますと胸壁の厚さにより多少の相異はありますが 5000~5500 r 位になります。現在の所外面照射のみによる胸部食道癌の治療には 5000 r を目標としております。

山下：1) 梶林君に対する回答

胸腹部のものでは淋巴節転移の分らないことがしばしばあり、それが予後に大いに関係すると思います。

2) 肺線維症に対する附議回答

食道癌に対する放射線療法では肺線維症はほとんど見ていない。それは従来照射線量が少なかったためかも知れない。回転照射で照射中心をはっきり定め、照射中心を細長くしてやると、もっと線量を増しても十分に肺線維症は防止できると考えます。

3) 食道癌の放射線治療成績はまだ非常に不良で、これをいかにして向上させるべきかであります。最も慎むべきは過量照射で、最適線量が分りつつあるように考えます。

216. RADIATION THERAPY OF ADVANCED STOMACH CANCER

HIDEO IRIE, KEIICHIRO ONIZUKA, TERUZO YASUDA

and ATSUSI KASAHARA

(Dept. of Radiol., Faculty of Med., Kyushu Univ.)

Radiation therapy of stomach cancer has been hitherto considered not effective.

In the author's opinion, if the technic is adequate, the radiation therapy is the effective method for advanced, inoperable stomach cancers.

The authors performed the radiation therapy, carefully devising the dosage, field size and interval according to the condition and reaction of the patients.

The results from 1949 to 1952 are as follows,

	number of cases	3 yr survivals
inoperable, irradiated	24	2
inoperable, not irradiated	25	0
recurrence after operation, irradiated	23	0

附 議

橋本和之：術後放射の価値は今改めて検討する意義がある。示説 217 におけるその成績を御覧いただけましたか。

入江：演者の考え方に賛成していただいて有難い。なお手術前のいわゆる Vorbestrahlung も同じような考え方で効果があるものと思う。

山下久雄：胃癌の放射線治療成績は非常に悪く、従来はむしろ照射すると悪くなるようにさえ考えられていたのに、Co-60 大量照射法などが広く行われるようになって、入江教授が述べられたように、相当進行した癌に対しても効果があり、また橋本教授がいわれたように術後照射の効果も少くないことが分つてきたわけで、広く皆様の御追試を願いたい。

217. FOLLOW-UP STUDY ON 582 CASES OF GASTRIC CANCER IRRADIATED AFTER SURGICAL OPERATION

KAZUYUKI NARABAYASHI, SHUJI KIMURA, KAZUNORI MAEDA,

HIROSHI MAENISHI, MIYAKO MANABE, SHINGI ITO

and YO IZUMI

(Dept. of Radiol., Kobe Med. College)

The patients were irradiated in our clinic from 1952 to 1959, following resection of the tumors. Result of 185 cases could not be obtained.

In 397 cases of gastric cancer, 139 patients had radical operation. Its three- and

five-years survival rates showed 66.8 per cent and 52.1 per cent, respectively, the number of which were higher than that by surgical operation only (25~45 per cent). The number of the survivals by postoperative irradiation was impressed to increase proportionally to the tumor doses given.

Our suggestion is that the higher survival rate of gastric cancer may be obtained by the improvement of the radiation method.

218. STUDY ON THE RADIATION THERAPY OF CARCINOMA OF THE CERVIX UTERI

OSAMU KOJIMA and MYOHEI WATANABE

(Dept. of Obst. & Gynec., School of Medicine Kumamoto Univ.)

Our technique of radiation therapy for the carcinoma of the cervix uteri consists of intra cavitory irradiation with ^{60}Co sticks against the primary lesions and ^{60}Co teletherapy against the parametrium with pelvic lymph nodes. The amount of dosage delivered to point A reaches at about from 6000 r to 8000 r and to point B, at about from 3500 r to 5000 r. Apparatus of ^{60}Co teletherapy which we have utilized is model RI-103 C (250 curie) made by Toshiba Electric Corporation.

Our principal method of radiation has been consisted of internal and external ^{60}Co therapy for more than two and half years since January 1958. Therefore, five years survival rate has not been able to be evaluated, however, one or two years survival rate was calculated. One year survival rate in 97 cases was 81.4%, and that of two years in 45 cases, 60.0%.

The classification of stages predicted their prognosis satisfactorily. Favourable results were obtained in patients whose histological specimens indicated moderately differentiated squamous cell carcinoma. Seventy mortality rate was estimated in 10 cases of 40 years old or younger. Prognosis depended upon clinical response, that is, good clinical response indicated good prognosis and the patients who had the poor clinical response, had all died. Complication of local infection induced poor prognosis, but rectal injury by irradiation had no distinct relation to prognosis in our series.

The presence of sensitization response in the basal cell of the vaginal smear was associated with two years survival rate of 72.4%, while its absence was associated with two years mortality rate of 66.0%. Furthermore, we have investigated the cause of death and reported the cases of radioresistant.

Finally, above mentioned survival rate of radiation therapy is still unsatisfactory and needs further discuss upon the method or radiation dose.

附 議

山下久雄：Manchester 方式による腔内照射と体外からの Co-60 照射とを併用して行う場合には，Wedge filter などを使用して A 点の線量が過重にならないような方式がとられることが望ましい。

小島：腔内近接照射は ^{60}Co 小線源を用い 3500~4500mch を照射をなし。さらに遠隔照射法で不足線量を追加するすなわち A 点に 2400 r, B 点に 3600 r 以上を照射する方針をとっている。

219. CLINICAL EXPERIENCES OF PLASTBALT-MACRO IN CASES OF CANCER OF UTERINE CERVIX

HAYAMI FUJIMORI, FUMIO YAMADA, SHIGEKAZU MAEDA,
HIROSHI KINOSHITA and MASATAKA MORIMURA
(Dept. of Obst. & Gyne., School of Med., Osaka City Univ.)

Plastobalt-Macro was used for the treatment of the cancer of uterine cervix.

Improving findings were recognized during and after the irradiation, by ordinary examination, by histological and cytological examination and by investigation with colposcope. Subjective complaints, for instance flow or bleeding, decreased by the irradiation of about 6000 mch. No severe complications were occurred during and after the treatment, and it might be concluded that plastobalt-Macro was suitable for the local irradiation for inoperable cases of the uterine cancer, because of the superiority of variable methods of application.

X. Therapy

220. COMBINED SURGICAL RADIATIVE TREATMENT FOR CANCEROUS LESIONS WITH PARTICULAR INTEREST IN PREVENTION OF RECURRENCE (THEORETICAL BASIS FOR THE PREOPERATIVE IRRADIATION IN THE TREATMENT OF ESOPHAGEAL CANCER)

KOMEI NAKAYAMA, FUMINORI YANAGISAWA, YASUMASA HONMA,
TADAO KAMATA, KEINOSUKE SUZUKI, NOBUJIRO TAKIZAWA
and HIROTAKE KAKEI

(Dept. of Surgery (Nakayama), Dept. of Path., Dept. of Radiol., Chiba Univ.)

Operative mortality rate for the treatment of the esophageal carcinoma is generally high, however, we have accomplished performing these operations in 1161 cases with operative mortality of 6.0%, which has become relatively easy and safe procedure. Their 5 year survival rate is 13.9% in 107 cases operated during the years of 1946 to 1953, which is better than those with simple irradiation, (Smithers 1936-1951, 3.9% in 229 cases) for the treatment of the upper and mid thoracic esophageal cancer. However, this figure is very unsatisfactory when one compares this with the results obtained from surgery on cancer of the stomach, which is 23.8% (1946-1952). In order to improve this poor operative results, pre-operative irradiation was administered to 226 cases since 2 years ago, and 114 cases with cancer of the esophagus included. Purpose of this pre-operative irradiation is 1) to inhibit proliferation of carcinoma and spontaneous metastatic process, 2) prevention of manual contamination of cancer cells during surgery, 3) widening of the operative indications. Contra-indications for this preoperative irradiation are 1) worsening of the general condition, 2) to makes operative procedure more difficult, 3) to delay and miss proper time for the radical operative treatment. In order to fulfill some requirements for this process, such as optimal irradiation dosage and proper timing for surgery upon completion of the irradiation, histological pictures of various cancer cells following irradiation of various degrees were studied. For the purpose of classifying various histological findings, D, X, L, classification method was employed. "D" stands for differentiation of the malignant cells, "L" stands for infiltration of the tumor into different layers and "X" stands for changes of malignant cells due to irradiation. Each classified groups were graded into 3 according to their grade of malignancy.

X₁ is the histological picture with nuclear mitosis, alkali protoplasma and proliferation is observed, X₂ is the one with nuclear swelling and piknosis with vacuolar changes in cytoplasm. Definite changes due to irradiation are observed in entire picture. X₃ is the one with disintegration and loss of nucleus can be seen. This is the stage that the irradiation has clinical effects. Relation between this X grading and amount of irradiation which were actually used, was evaluated and after irradiation of between 2000 and 3000 r, histological changes into X₂ and X₃, i.e., inhibiting effect of cancer growth were observed. Even one month after the irradiation of large dosage, 6,000 r, proliferation of the cancer cells are observed in the cicatrix. Thus optimal irradiation dosages to the cancer of the esophagus is between 2,000 to 3,000 r and period of irradiation is about 10 to 14 days after completions of the pre-operative irradiation. Decrease in ³²P and staining grade in macro-radio-autograph were observed. Ratio of phosphor fragmentation and radio isotopes in cancerous tissues are decreased as well as RNA and DNA. There are considerable differences in metastatic lymph nodes. There is no case in which this irradiation made the operation impossible. Radiographic X ray defects of longer than 8 cm were compared with irradiated group and non-irradiated group, and resectability of the lesions were doubled among the radiated group, and their post-operative survival rate have increased. 53 cases of 2 yr. follow-up study for the cases with irradiation, reveals 50% and large improvement is observed, compared with the cases with operation only or irradiation only.

附 議

明石勝英：大変興味深く拝聴しました。本邦においては子宮癌根治手術前の放射線の利用はほとんど行なわれていない。私は2年前より約100例の子宮頸癌にラジウム 4000~5000 mg/st 前照射してより約10~14日後に手術している。演者の胃癌手術における Data のごとく満足すべきものであった。ラジウム+X線深部照射後に行える若干例もあるが、大体同様なことがいえる。永久治療の問題はまた別個であるが照射前手術不能とせられたものも可能となる例にしばしば遭遇する。原腫瘍の縮小と感染の消退によると考える。

山下久雄：非常に興味深く拝聴しました。われわれ放射線科医の立場としては、たとえ術前照射とはいえ、5000 r を照射していただきたい。そうすれば、さらに照射の効果は大きくなり、しかも、そのために手術を困難とすることはないと考えています。

221. HISTOPATHOLOGICAL EVALUATION OF CANCER OF THE LOWER ESOPHAGUS AND CARDIAC END OF THE STOMACH

TOSHIO ITO, FUMINORI YANAGISAWA, YASUMASA HONMA,
KEINOSUKE SUZUKI, MICHIO IWATSUKA, HIROTAKE SUZUKI
and SHUN'ICHI TAMAMA (Dept. of Nakayama Surg., Chiba Univ.)

Cancer of the lower esophagus and of the cardia is still considered as one of the

most difficult lesions that can be treated surgically. There are less than 10 reports in the literature who have experiences in operating more than 100 cases. 841 cases of radical removal of the lesions with operative mortality of 4.7% have been carried out. Follow up results of these radically treated cases are, however, unsatisfactory and their 5 year survival rate is only 11.6%. Histopathological evaluation of 195 removed specimen were done with particular interest into the long survived cases. Cases who died within one year after the operation, showed no significant differences from those who survived in macroscopic inspection. 22 cases out of 31 cases who survived longer than 5 years (70.9%) are of circumscribed type only and 9 cases are infiltrative type. In circumscription of the tumor in infiltrative type is so poor that the possibility of leaving the tumor infiltration beyond the site of amputation is great. Aggressive thoracotomy and inspection of the entire tumor is recommended since recurrence at the site of anastomosis is seen more frequently in cases with abdominal approach. 39 cases out of 195 have diameter of shorter than 5 centimeters in their gross tumor (20%) and their 5 years survival rate is 36.6%. Survival rate is 17.4% among the group with its diameter between 5 and 10 centimeters and 4.7% among those whose tumor's diameter is longer than 10 centimeters.

60.5% of the entire cases showed tumor infiltration into the serosal layer and its 5 yr. survival rate is 12.5% and those with tumor localized within the submucosal and muscular layer has survival rate of 24.1%. Lymphatic metastases are seen in 68.1% of the entire cases and the group with metastases showed 11.0% in 5 yr. survival rate and 27.6% in non-metastatic group. Considerable numbers of patients who survived with apparent lymphatic metastases, probably means that thorough removal of lymphatic channels and adjacent organs with the primary tumor are one of the most important element to obtain longer survival after operations. Considerable differences are seen in histological pictures between the primary tumors and lymphatic metastases and this makes very difficult to grade the malignancy of the individual tumor. Out of entire 195 cases evaluated histologically, squamous epithelial group was 36, cylindrical epithelial 106 and carcinoma simplex was 53. 5 yr. survival rate of these groups are, 15.9% 13.2% and 11.3% each, and carcinoma simplex is the worst as expected. These results were further evaluated as to the maturity of their cells and well differentiated squamous epithelial 22.8%, well differentiated cylindrical epithelial 22.0%, undifferentiated squamous epithelial 14.3%, undifferentiated cylindrical epithelial 10.5% and carcinoma simplex 11.3% in these long survived cases. Undifferentiated cancers showed almost same results as to the ca. simplex and well differentiated groups showed a good operative results. (19 cases out of 31 who survived more than 5 years.) Cancer of the lower esophagus and cardiac end of the stomach loses their adequate time for early operation in radical cure if their grade of malignancy is high. Thus from surgeon's point of

view, the earlier diagnosis and earlier operation is recommended to improve radical cure rate for the treatment of the cancer of lower esophagus and cardiac end of the stomach.

222. CANCER OF THE STOMACH —AN ANALYSIS OF 2343 CASES—

TAMAKI KAJITANI, TOMOO HOSHINO, KUNIO TAKAGI
and MAKOTO NAKAMURA

(Dept. of Surgery, Cancer Institute Hospital)

A total of 2383 cases of gastric cancer were admitted to the Department of Surgery in the Hospital of Cancer Institute from Sep. 1946 to Dec. 1959. This is a report of 2343 cases which underwent operative procedures.

1. The average age of 2343 patients was 53.6 years. The youngest was 16 years and the oldest 80 years. 1523 (65.0%) were male and 820 (35.0%) were female.

2. Heavy feeling in the stomach was the first symptom most frequently noticed. This complaint was noted in 44.3% of 1810 patients (1952-1959). Pain was noted in 42.8%, fullness 39.3%, anorexia 25.5%, loss of weight 16.5%, belching 15.6% and heartburn 13.7%.

The types of symptom at the initial stadium of gastric cancer except cancer of the cardia (156 cases) were as follows: (a) indefinite symptom type.....1210 (73.1%), (b) ulcer-like symptom type.....318 (19.2%), (c) extragastric symptom type.....105 (6.4%), (d) fulminant type.....21 (1.3%).

3. Achlorhydria was noted in 1357 (59.9%) of 2264 patients tested, hypochlorhydria 420 (18.3%), normal 329 (14.5%) and hyperchlorhydria 158 (7.0%).

4. Curative resection was performed on 1495 of 2343 patients (curative operability rate was 63.8%) and 47 died within one month (operative mortality rate was 3.1%).

In 1495 patients, total gastrectomy was performed for 145 patients, total gastrectomy with resection of adjacent organs for 285 patients, proximal gastrectomy for 5, proximal gastrectomy with resection of adjacent organs for 6, distal gastrectomy for 872 and distal gastrectomy with resection of adjacent organs for 182.

5. The most important condition which precluded the curative operation was the peritoneal seeding, which was noted in 344 of 848 patients who underwent palliative procedures (40.6%). Peritoneal seeding with hepatic metastasis was present in (6.0%), hepatic metastasis in 154 (18.2%), nonresectable lymph node involvement in 242 (28.5%) and continuous involvement in 231 (27.2%).

6. Two hundred and sixty-one in 659 patients who underwent curative resection

from 1946 through 1954 survived 5 years or longer (five-year survival rate was 39.6%).

(1) Macroscopic type of cancer has a great relation to survival rate. One hundred and thirty-nine of 248 localized type survived 5 years (56.0%), while 122 of 411 nonlocalized type survived 5 years (29.7%).

(2) Number of lymph node positive cases were 493 (74.8%) and 29.6% of those cases survived 5 years. In contradiction, the five-year survival rate of 166 lymph node negative cases was 69.3%, especially in the localized type of cancer it was noted as high as 76.2%.

(3) In the nonlocalized type of cancer, invasion into serosa of the stomach has close connection with the survival rate. In 94 cases in which serosal invasion was negligible, 49 (52.1%) survived 5 years. Of 119 cases with narrow invasion, 42 (35.2%) survived 5 years and of 138 with moderate invasion, 30 (21.7%), but of 60 cases with wide invasion only 1 (1.7%).

(4) As to the histology, the five-year survival rate of the medullary cancer was 44.9% (193 in 430), gelatinous cancer 50.0% (22 in 44), scirrhous cancer 24.7% (45 in 182) and squamous cell cancer 33.3% (1 in 3).

(5) Correlation of the survival rate to the grade of malignancy (Broders) was as follows: Grade 1; none, Grade 2; 48.0% (74 in 154), Grade 3; 42.8 (115 in 269) and Grade 4; 30.5% (72 in 236).

附 議

陣内：胃癌根治手術後5年生存率 39.6% という大変立派な御成績をみせていただきましたが、私方の成績は毎年の統計で多少変わりますが 34.8~39.1% で少し劣ります。鳥取大学綾部外科は 40% という成績であります。これは取扱う材料にもよるわけで、御承知のごとく綾部君は初期癌の大家ですので、潰瘍、胃炎の胃を切除して組織学的に検索して初期癌を発見したものも含まれているようですが、とにかくも今日 40% 近くに成績が向上したということは、やはり努力して根気強くリンパ節廓清を行うことが確かに有意義であることを示すものと存じます。

まだまだ外科の手術は眼科、耳鼻科の手術に比べると技術的に grob でありまして、外国では脳外科などでは拡大鏡をかけてやっている人があります。将来癌の外科もこのように精密な外科技術に向ってゆくものと信じております。

223. THE CARCINOMA DEVELOPMENT IN THE GASTRIC STUMP AFTER PARTIAL GASTRECTOMY FOR BENIGN GASTRODUODENAL LESION

MASANOBU TOMODA and MASAZUMI MIZOKUCHI

(2nd Dept. of Surgery, Faculty of Med., Kyushu Univ.)

Carcinoma developing in the stump of gastric remnant some years after partial gastrectomy for benign gastroduodenal lesion (gastric stump carcinoma) has been

recently drawing attention especially in Europe and America. Report on such cases has been however very seldom in Japan.

The problem of gastric stump carcinoma will need more attention and reevaluation in Japan as well, while partial gastrectomy for benign lesion of the stomach or duodenum is now frequently employed and there are increasing numbers of partial gastrectomy against precancerous lesion of the stomach in view of prevention of gastric carcinoma.

Our recent experiences with 2 cases of gastric stump carcinoma after partial gastrectomy are described. One (microtubular adenocarcinoma) developed 20 yrs. after operation for gastric ptosis and the other (medullary carcinoma) developed 19 yrs. after surgery for gastric and duodenal ulcers.

Regarding these 2 cases the problem of gastric stump carcinoma is pathologically and surgically reviewed.

附 議

今井 環：このような癌が、「断端」そのものに初発したものか、あるいは残胃のどこかに生じて、断端線に達したものか、そのいずれであるかによって、その原因や対策にも異なるところが出てくるのではないと思われるが、もし、語義通りの断端癌だとすれば、そのように判定する基準(根拠)は、どう定められておるか、お教示いただきたい。

溝口：断端癌は胃腸吻合部の胃粘膜に原発したものをいい、残胃に胃腸吻合部とは無関係に発生した胃癌とは別である。

断端癌の初期像の経験はないので厳密にはいえないが、経験した2例の肉眼的および組織検査では、吻合部胃粘膜に原発したと思われる胃癌の所見を呈していた。

村上忠重：長年月前に良性潰瘍のために胃空腸吻合術(胃切除を行わない)を施行された患者の胃に胃癌の発生した例を2例経験している。1例は潰瘍癌で、おそらく残留した潰瘍を母地としたものと思われる。他は胃空腸吻合孔に一致したドーナツ型の癌で、吻合のために胃に加えられた創がそのまま発癌の場となったのであろうと推定された。したがって断端癌という言葉があつてよいと考える。

友田：演者の講演に一寸補足申します、胃十二指腸良性疾患に対する胃切除後に胃断端癌が発生するとして、この問題は発癌、胃切除の治療的意義等の点を中心として主要な研究問題をふくんでいます。したがって今後かかる症例の発見についてとくに内科医の関心を希望いたしますと存じます。

発生部位についての今井教授の御質問に対しましては、「断端癌」は胃腸吻合部に接した胃断端に発生したものと考えられます。外科にきますときは、相当進行していますから、発生の初発部位を決定することはむずかしいですが、占居部位から、上記のごとく判断せられます。断端から相当離れて生じたものが、経過中断端に波及したものとは考えられません。

224. THE PROBLEM OF PANCREATIC LYMPH NODES IN GASTRIC CANCER SURGERY

**TOSHIO OIWA, TOMOHIRO TODA, HIRAKU SAITO
and YUKIAKI HARAGUCHI**

(2nd Dept. of Surg., Fac. of Med., Kyushu Univ.)

The relation of the stomach with lymph nodes existing in the neighbourhood of the pancreas was examined from the surgical view point. Further, the metastasis of gastric cancer cells to these nodes were studied.

In 64 fresh human fetuses the stomach was classified into three parts; proximal, middle and distal part. India ink was injected into subserosa of the three parts. It was found that the region of india ink inflow in lymph nodes varied depending upon the site of injection in the stomach. Splenic hilus and suprapancreatic lymph nodes were closely related with proximal portion of the stomach. Distal part of the stomach was related with subpyloric, hepatic and mesenteric root lymph nodes. Coeliac lymph nodes were found related with every parts of the stomach.

The local relationship of suprapancreatic nodes with splenic vessels was histologically examined in 10 autoptic cases. In most cases suprapancreatic nodes existed on the splenic vessels, and in some cases lymph nodes existed in between splenic artery and vein. This part deserved attention because there was high incidence of metastasis when proximal part of the stomach is involved and there were also some cases where cancer cells were found in lymph vessels.

Lymph nodes surgically dissected from 72 cases of gastric cancer were histologically studied. The results were correlated with our basic finding and the importance of dissection of lymph nodes around the pancreas in the gastric cancer surgery is stressed.

225. CLINICAL OBSERVATION OF 75 CASES OF PRIMARY CANCER OF THE LUNG IN OUR CLINIC IN THE LAST 5 YEARS

**YOSHIO HARA, KAZUO KATSUTA, MINORU TAKAHASHI,
BUNYA ITO and TSUNEO HOSHINO**

(Dept. of Med., School of Med., Niigata Univ.)

Incidence of the primary lung cancer in all patients has increased from 0.20 to 0.37% and incidence of it in all cancers from 7.5 to 14.9% in the last 5 years (1951

to 1960) in our clinic. 1) In our 75 cases 57 were male. 2) Peaks were seen in 5th decade in male, and 6th in female. 3) Hereditary relationships were not dominant. 4) Relationship to tobacco was definite. In 52 male patients 26 smoked more than 20 cigarettes per day. 5) Frequent initial symptoms were cough (5.6%), sputum (21%), chest & back pain (20%), bloody sputum (16%) and dyspnea (7%), but 15 cases had no symptom when diagnosis was first made on survey chest X-ray. 6) Diagnosis made by first seen doctors were pulmonary tuberculosis 14, common cold 8, intercostal neuralgia 6, pleurisy 6, bronchial asthma 4 and bronchitis 2. 7) B.S.R. were more than 20 mm/hr. in 52 out of 68. 8) Hb. were more than 80% in 21 out of 36 male patients and more than 70% in 11 out of 24 female patients on admission. 9) In 13 out of 75 cases pleural effusion was seen on admission and 6 were in the right side, 3 were in the left side and 3 were seen bilaterally. 10) Bronchoscopic examinations revealed positive findings in 18 out of 23 cases. 11) Tumor cells in pleural exudates were found in 13 out of 17 cases. 12) Localizations of primary lesions were most frequent in the right upper lobe, second in right lower lobe and third in left upper lobe. 13) Histologically 7 were epidermoid cell ca., 8 were anaplastic and 4 were adenocarcinoma. 14) 3 in 75 cases complicated with pulmonary tuberculosis. 15) Average duration of life of 43 died cases was 9.1 months. 16) Our all cases were treated with some anti-tumor drugs, but their average lives were shorter than other reports.

附 議

宮地 徹：御発表のなかで、組織学的に扁平上皮癌と未分化癌が多いということでしたが、これは全例について組織学的検索を行った結果でしょうか。(剖検は 18 例で、それについてののみ組織学的検査を行ったとの返事があった)。それでは、組織像の頻度は大きい例数を取扱った場合とは差が出るのは当然と存じますので、この点にはあまりウエイトをおかけになりませんようにお願いいたしたく存じます。

河合正次：内科的に観察された臨床症状についての御報告と思うが、外科の経験からいうと三段階があると思う、末期、中間期、早期とに大別されるが、これによって症状とか生存日数などいちじるしい差違があるのは当然であるから早期発見、早期治療を第一の目標としている現況では、これに注目して検討していただきたいと思う。

226. THE RESULT OF SURGERY OF ADRENALS FOR ADVANCED CANCER OF THE BREAST

RYOJI NAGAI, MITSUHIKO SENGOKU, MINORU YOSHIDA
and MITSUO UKAI

(Imanaga Surgical Clinic, School of Med., Nagoya Univ.)

The sexual hormones play an important role in the development and the metastasis of breast cancer, and a part of them is excreted from the adrenal cortex. In our

clinic, adrenal surgery has been done for the treatment of advanced cancer of the breast. Since November 1956, forty patients have been submitted to adrenal surgery. In twenty-one cases, the portalization of adrenal venous blood with exstirpation of the right adrenal gland and bilateral ovaries was performed, and in seven cases, the bilateral total adrenalectomy and oophorectomy. Objective improvement was observed in about half of the cases so treated.

1. Result of Adrenal Surgery

	Evaluated Cases	Objective Improvement	
		Effective	None
Portalized Procedure	20	11 (55%)	9 (45%)
Total Adrenalectomy	16	9 (56%)	7 (44%)
Total	36	20 (56%)	16 (44%)

2. Mean Survival Time after Adrenal Surgery

	Alive	Dead	Total
Length of Survival (Months)			
Effective Cases	19.6	8.4	14.1
None Effective Cases	1.0	2.9	2.8

3. Mean Time of Objective Regression after Adrenal Surgery (Effective Cases)

	Regression Ceased	Now in Regression	Total
Length of Regression (Months)	9.7	11.2	10.1

4. Objective Symptoms Improved

	Total Cases	Effective Cases
Diminution of Metastatic Tumors	36	19
Calcification of Bone Metastasis	9	4
Epithelization of Ulcerative Cancer	5	5
Disappearance of Edema	12	6
Disappearance of Cranial Symptoms	2	1

It is concluded that the adrenal surgery is one of the most preferable treatment for advanced breast cancer.

227. ANNUAL REPORT OF THE JAPANESE COMMITTEE OF CARCINOMA OF THE UTERUS (II)

HIDEO YAGI (The Japanese Obst. and Gyne. Society)

Before the 18. General Meeting of this Cancer Association 1959, I read the first report of the Committee on the results of treatment of carcinoma of the cervix collected from 21 institutions throughout Japan, which contained a five year cure of 1,806 cases treated in 1953. To this I can add the five year results of 2,447 cases of carcinoma of the cervix reported by the same 21 institutions and 12 others as new

members, thus making cumulative statistics of 4,253 cases for 1953 and 1954.

The distribution of clinical stages was: Stage I 898 (21.1%), Stage II 1,629 (38.3%), Stage III 1,334 (31.4%) and Stage IV 392 (9.2%). The five year cure was 48.9% on the average (Stages I-IV). According to stages, it was 78.6% for Stage I, 56.5% for Stage II, 31.2% for Stage III and 9.7% for Stage IV.

If the total cases were divided into two groups, surgery and radiation, the cure rate was as follows: (1) surgical group: 83.1% for Stage I, 62.0% for Stage II, 44.8% for Stage III and 7.1% for Stage IV, while (2) radiation group: 55.2% for Stage I, 42.7% for Stage II, 24.9% for Stage III and 9.8% for Stage IV. Thus the results of radical surgery were better for all stages except IV than radiation.

附 議

山下久雄: 従来わが国においては、子宮癌の放射線療法に関して婦人科医と放射線科医との間に意志の疎通をかねていたことを痛感するものであります。

40年前のラジウム療法で子宮癌はある程度治癒しますが、いまだに I Serie, II Serien, III Serien という照射術式をとっておられる日本の多くの婦人科の照射術式ではこれ以上の向上は望めない。欧米に劣るのは当然の結果だと思います。

放射線療法は日進月歩がいちじるしいのですから、今後は放射線科医と密接な連絡をとられて、欧米に劣らない治療成績を挙げられるように希望するものであります。

まず第一にやっていただきたいことは、(1) A 点に対しては腔内照射で十分の線量を与え、外部照射は Wedge filter で遮断すること、(2) 外部照射は B 点に主眼点を置いて、最初の機会に治して了うだけの十分の線量を与えることにあてると考えます。

重松 康: (1) 日本の放射線治療成績が外科手術より非常に低いということは、放射線治療医と婦人科医との協力のあり方の悪さに最大の原因があると思う。

(2) 殊に放射線治療に関しては照射方式を十分に吟味してみないと批判の材料とはなり難い。

(3) 少くとも照射術式の選択改善で欧米の成績までは引上げ得ると信ずる。

(4) この問題に関しては山下博士に発言を求める。

八木: 放射療法が手術成績に比し悪いことの一つにわが国における放射器械の性能が必ずしも全部は優秀でないことも一因であるが、他面放射術式、X 線のかけ方、Ra の入れ方、Co の用い方、これらの合併方法等の因子も大きく作用する。これらには是非婦人科医と放射専門家との協力が望ましい。岡大では常に協力して治療をしている。I Serie, II Serie のやり方はすでに廃止し I Serie のみに十分な線量を与えることにしている。

手術の方式、放射の方式の詳細については時間がなく申し上げられないが、委員会としては優秀な成績を挙げた法式を示して、他がなるべくこれに追従してもらうようにし、日本全体のレベルを向上せしめるように努力するつもりである。

小林 隆: 日本において子宮頸癌に対する放射線療法の成績と手術療法のそれとの差が余りにも大きいことの理由の一つには症例の選択やかたよりが考えられないことはない。すなわち日本では放射線療法には poor risk 的なものおよび進行したものが集まりやすく、手術療法にはその逆のものが多くなりやすいからである。それはともかくとして日本の手術療法の成績が外国の最良のそれにくらべてなおまざっていることが日本ではメスを主流とさせている理由であってこれが同じになれば放射線療法がこれにかかわることは疑い得ない。しかし次のことに留意する必要がある。すなわち放射線の侵襲のメカニズムは一つ一つの細胞のレベルであるのにその治療成績は優劣はとにかくとして少くとも手術療法のそれと近似するのはどういふわけであろうか、換言すると手術療法では第 1 期第 2 期の間に成績の差を生じるのは当然と思うが放射線療法では細胞レベルで侵襲する以上差を生じないか少くともその差はいちじるしく小さくなるはずではないか、事実は第 1 期と第 2 期との間に手術と同じような差を生じている。

228. CARCINOMA OF THE CERVICAL STUMP

KAZUMASA MASUBUCHI, HISAMITSU KUBO and TADAO SUZUKI

(Dept. of Gyne., Cancer Institute Hospital)

2291 cases of carcinoma of the cervix including 146 cases of carcinoma *in situ* were treated during 1950-1959.

- 1) Fourty four cases (1.9%) were carcinoma of the stump.
- 2) Thirty five cases (79.5%) of them had been performed the subtotal hysterectomy because of myoma uteri.
- 3) The clinical staging: stage 0, 2 cases (4.5%); stage I, 11 cases (25.0%); stage II, 18 cases (49.9); stage III, 12 cases (27.3%) and stage IV, 1 case (2.3%).
- 4) The age distribution: 30 to 39 years old-6.8%; 40 to 49-43.2%; 50 to 59-45.5%; and 60 to 69-4.5%.
- 5) The average gravidity: 2.9 in carcinoma of the stump, 5.7 in carcinoma of the cervix. The average parity: 2.5 in carcinoma of the stump, 4.1 in carcinoma of the cervix. Sterility: 11.4% in carcinoma of the stump, 5.6% in carcinoma of the cervix. Nullipara: 15.9% in carcinoma of the stump, 7.2% in carcinoma of the cervix.
- 6) In most cases (86.4%) vaginal bleeding was the initial symptom in carcinoma of the stump.
- 7) The incidence of adenocarcinoma was higher in cases of carcinoma of the stump (13.6%) than in carcinoma of the cervix (5.9%).
- 8) From the standpoint of treatment, both operation and irradiation are slightly less advantageous in stump cancer.
- 9) Five years survival were 3 cases out of 5 cases in stage I, 7 out of 7 in stage II, and 1 out of 5 in stage III.
- 10) The interval between subtotal hysterectomy and diagnosis of carcinoma: 1-5 years 18 cases (40.9%), 5-10 years 6 cases (13.6%), over 10 years 20 cases (45.5%).
- 11) There were additional 7 cases which were detected carcinoma at the stump within one year following subtotal hysterectomy.
- 12) Really, it is very difficult to differentiate the stump cancer from recidual cancer at the stump, but from the fact that there were 25 cases in which carcinoma was detected within 5 years following subtotal hysterectomy, it would be the last chance to avoid the development of stump cancer, if the total hysterectomy had been performed instead of subtotal one.

附 註

増淵一正: 1) 腔上部切断術を受けた婦人はおおむね子宮の残存を知らされていない。断端癌の発生の危険を知らされていない。術者は当然退院時に説明しておくべきだ。

2) 本日の報告から判るように、特別の理由のないかぎり、腔上部切断術は廃止さるべき術式であると考えている。

小林 隆：筋腫手術のさいすでに潜在した子宮頸癌の問題はその病院での Cancer Detection の方針の精粗や能力に関係すると思いますが一方筋腫手術後の子宮断端の発癌性そのものについてはいかにお考えですか。手術によって増強されるかあるいは全く影響ないものかなどについてうかがいたい。

久保：5 年治癒例が 17 例にすぎませんので症例追加をまって改めて御報告いたしたいと思います。

229. STUDIES ON THE VAGINAL SYSTEMATIC RADICAL HYSTERECTOMY FOR CANCER OF THE UTERINE CERVIX AND THE EXTRAPERITONEAL PROCEDURE OF LYMPHNODECTOMY WITH LIGATION OF THE PELVIC VESSELS (II)

KATSUhide AKASHI, MASAYOSHI HASHIMOTO, AKIRA KOMORI
and YUTAKA YAMADA

(Dept. of Obst. & Gynec. Sapporo Med. College)

The vaginal radical hysterectomy for cancer of the uterine cervix without lymph-nodectomy is considered to be an incomplete method. The present report is presented to give a more systematic radical modification to the extraperitoneal procedure of pelvic lymphnodectomy with ligation of pelvic vessels and the vaginal procedure.

Material and Method of Operation.

This procedure was performed from August 1957 to November 1960, 150 cases of uterine cervical cancer (Stage I, II) at the Department of Obstetrics and Gynecology, Sapporo Medical College, Hokkaido, Japan.

I. Extraperitoneal procedure following:

- 1) Median section of abdominal skin.
- 2) Extraperitoneal separation of pelvic lateral cavity and dissection of the vessels for blood sparing:

A. et V. epigastrica inf, Lig. teres, V. Plexus pampiniformis (ligation), A. iliaca int. (ligation), A. et V. obturatoria and A. uterina.

3) Extraperitoneal lymphnodectomy:

Lnn. inguinale profunda, iliaca ext. inf. et superior, interiliaca, comm. lat. (partial), obturatoria, parametran, and paravesicalis etc. are dissected as a continuous tissue-lamina, as possible as, with perivascular tissue and fat tissue.

- 4) Abdominal section is closed with gummidrainage.
- II. Vaginal systematic radical hysterectomy:
Vaginal operation are made immediately after the extraperitoneal procedure
 - 1) Formation of the vaginal cuff.
 - 2) Separation of the Bladder.
 - 3) A left Schuchardt's vaginoperineal incision.
 - 4) Transversal dissection at the bladderpillar and freeing of the ureter.
 - 5) Incision of pelvic peritoneum and the dissection of the adnexe.
 - 6) Closure of pelvic peritoneum with a running suture for easy separation of the rectalpillars and the cardinal ligaments.
 - 7) Dissection of rectal pillars.
 - 8) The separation of the cardinal ligaments at its lateral settled portion and the dissection as the thin band (sog. V. cardinalis).
 - 9) Gummidrain in bilateral fossa iliaca per vaginale.

Result :

I) The characteristics of this operation are 1) the extraperitoneal lymphnodectomy with ligation of the pelvic vessels and 2) vaginal systematic radical hysterectomy; horizontal dissection of bladderpillars to be made free of the ureter and the closure of peritoneum, immediately after the adnektomy, for the easiness to broaden the separation and dissection of the rectal pillars and the cardinal ligaments.

II) Primary mortality;	0.8%
III) Morbidity;	20 %
IV) Uretero-vaginofistel;	2.8%
V) Bleeding volume;	avarage 750 cc

230. CLINICAL AND PATHOLOGICAL STUDIES ON THE KRUKENBERG'S TUMOR

KAZUSHIGE HIGUCHI, TOSHI KATO, TARO TAZAWA, SHIGEO OGAWA,
TERUO KOBAYASHI, KIYOSHI HASUDA, SHIGETAKA KOBAYASHI,
YOSHITERU TERASHIMA, AKIRA TANAKA and HIDEAKI NAGUMO

(Dept. of Gynec. and Obst., Jikeikai Med. School)

A total 205 cases of Krukenberg's tumors (20.5%) was found among 1000 ovarian tumors collected in our department.

Statistic figures here are remarkably higher than those found in Europe and in U.S.A. The past history revealed diseases of the digestive system in 30.9% of the cases, and the chief complaints consisted of tumor of the lower abdomen and pain.

The ages of patient ranges between 17 to 65 years with average 35 years. About 66.8% of the cases were in their 30 and 40 years of age. Ascites (serous) was found in 71.0%. The size of tumors ranged from hen egg up to adult head sized.

Histologically, tumor cells metastasizing to the uterus were found in 76.4% of the cases. The tumor cells which were transported lymphatically from the primary origin to mesosalpinx and enter into lymphatics of the uterin horn and form metastases in the myometrium of the uterine body and portio.

Grossly, in the cases with normal appearing ovary on one side and gross metastatic lesion of the other demonstrate lymphatic metastases in 83.3% in the normal appearing ovary when closely examined. It may be stated that this type of tumor is bilateral and if not carefully examined there is possibility of detecting tumor cells in only one ovary. Three dimensional histological examination of tumor revealed adenocarcinoma 43.8%, solid carcinoma in 34.0% and sarcoma like lesions in 80.4%, scirrhous type in 77.1%, signet cell type in 85.2%. A combination of various types were often found in the same tumor.

(文部省科学研究費による)

231. FOLLOW UP STUDY ON THE TUMOR OF THE BLADDER

GOICHI MOMOSE and SHIN'ICHI MITSUHASHI

(Urolog. Clinic, School of Med., Chiba Univ.)

We carried out the follow up study of 129 cases diagnosed as bladder tumor coming to our clinic since 14 years and a half ago.

- 1) Prognosis is better in the cases having complaint of only hematuria than of any others such as having complaint of pollakisuria and pain at the micturition.
- 2) Mortality is higher in the cases having large tumor than small one.
- 3) Regarding the numbers of tumor, difference of the mortalities is not so remarkable as the size of tumor.
- 4) Mortality is distinctly better in the case without invading at orificium of ureter than others.
- 5) Prognosis of the case with advanced staging and grading is distinctly worse.
- 6) Prognosis is better in the case being microscopically papillomatous than non-papillomatous.
- 7) Better in the case of transitional cell tumor than in any others.
- 8) Remarkably worse in the case of intravascular tumor cell invasion.
- 9) Radical treatment is impossible when the dysfunction is in the kidney. Consequently prognosis is bad.
- 10) Prognosis is excellently good in the case performing the partial cystectomy

compared with the case of total cystectomy.

11) Total surviving rate during 5 years is very low at 35.6%.

附 議

小林 隆：膀胱癌は手術的にも放射線療法的にも子宮癌と酷似した条件を備えていると思うので今後の膀胱癌の治療の動向はわれわれ婦人科医にとっても興味深いものがあるわけです。 ^{60}Co やベータートロン^の出現が治療法や治癒率にどういふ動向や変化をもたらしつつあるか、あるいは将来の見通しは？

三橋：1) 治療の主流はある程度の再発を覚悟の上で、部分切除をすることがよい。

2) 全剝の場合の尿流変更は回腸膀胱がよいのではないか。

232. CLINICAL STUDY OF PRIMARY HYPERPARATHYROIDISM (PARATHYROID ADENOMA) IN UROLOGY

TAKAMITSU KUSUNOKI, TAKAO SONODA, TADASHI OHKAWA
and MASABUMI TAKEUCHI

(The Dept. of Urology, Med. School, Osaka Univ.)

Four cases of primary hyperparathyroidism due to adenoma of the parathyroid gland are presented. All these four cases revealed renal complications. This is the first report of clinically proved cases with urolithiasis due to primary hyperparathyroidism in Japan.

Since September 1958 to December 1960, 189 cases of urolithiasis have been treated in our clinic, and our four cases correspond to 2.1 per cent of these cases. This percentage is almost equal to that of the other statistics in western countries.

The diagnosis of primary hyperparathyroidism is rather difficult because of following reasons: the tumor of the parathyroid gland cannot be palpable throughout the neck, hypercalcemia, hypophosphatemia, hypercalciuria and hyperphosphaturia are not always specific complications of this disease and the serum level of parathyroid hormone has not been able to be estimated. From these points of view, we must wait for the accumulation of more accurate laboratory evidences, and for this purpose various laboratory tests such as urine concentration test, estimation of % TRP, phosphate deprivation test, calcium tolerance test, calculation of parathyroid index and estimation of theoretical renal phosphorus threshold (TRPT) are employed. We applied these tests to four cases of primary hyperparathyroidism and other control cases with or without urolithiasis, and the results were discussed. The simplified method for the estimation of theoretical renal phosphorus threshold was introduced.

From the results of these laboratory tests, we concluded that the following three stages for screening of the patient with primary hyperparathyroidism were necessary.

1. First Stage

- a) Clinical signs
 - b) Determination of serum calcium and phosphorus levels
 - c) Determination of urinary calcium and phosphorus excretions
2. Second Stage
- a) Urine concentration test
 - b) Estimation of % TRP
3. Third Stage
- a) Calcium tolerance test
 - b) Calculation of parathyroid index
 - c) Estimation of theoretical renal phosphorus threshold
-

XI. Growth

233. VARIATION IN ENZYMIC ACTIVITY WITH THE MITOTIC RATE

TAKASHI SHIMAZU and TOSHIHIKO SUEMATSU

(Institute for Cancer Research, Osaka Univ.)

During a study on the mechanism of metabolic control of liver regeneration after partial hepatectomy, investigations were made on various biochemical changes occurring in parallel with the increased rate of mitosis.

This paper reports studies on the aspartate-carbamyl transferase and deoxyribose phosphate aldolase of regenerating rat liver and rat hepatomas. The activity of aspartate-carbamyl transferase was measured in several rat hepatomas (including a DAB-induced primary hepatoma, ascites hepatoma, AH 130, and a solid hepatoma transplanted by inoculation of an ascites hepatoma into the portal vein). All had activities about 3 times higher than that of normal rat liver. In regenerating rat liver, the activity of these two enzymes after partial hepatectomy, varied approximately in parallel with the mitotic index, as judged histologically. The maximum activity of deoxyribose phosphate aldolase was about 2 times higher than that in normal liver, 36-48 hrs. after partial hepatectomy. The maximum activity of aspartate-carbamyl transferase was 2.5 times normal, 48-60 hrs. after partial hepatectomy. The activity of these two enzymes then gradually decreased to the normal level within 14 days after hepatectomy.

By fractionation of the subcellular constituents it was found that the aspartate-carbamyl transferase was only present in the cell-sap of normal rat liver but in regenerating rat liver and hepatoma, although most activity was found in the cell-sap, the microsomal fraction was also active. Deoxyribose phosphate aldolase was found only in the cell-sap fraction. Neither the nuclear nor the mitochondrial fraction had any enzymic activities with regard to these two enzymes.

Studies were made on the effect of the hormonal glands on rat liver regeneration after partial hepatectomy. The activity of the aspartate-carbamyl transferase was used as an index of the extent of regeneration. After either hypophysectomy, thyroidectomy, bilateral adrenalectomy or ovariectomy in rats (for these studies female rats were used), the liver was partially removed. The activity of aspartate-carbamyl transferase was assayed 52 hrs. after the partial hepatectomy. The results did not indicate any significant influence of hormonal glands upon the increase in activity

of this enzyme. It is thus unlikely that any of the hormonal glands studied in these experiments significantly affects liver regeneration.

Further studies on the effect of the autonomic centers (hypothalamus) on liver regeneration are now in progress.

234. STUDIES ON COLLAGEN FORMATION IN TISSUE CULTURE
(III) ON THE COLLAGEN FORMATION BY THE STRAIN
CELLS (JTC-4 AND -6) ORIGINATED IN RATS

RYOSABURO TAKAKI, YOKICHI SUGI, KOOICHI TAKANO*,
KIMIE KANEKO*, MAKOTO UMEDA**, TOSHIKO TAKAOKA**,
HAJIM KATSUTA**, HIROYOSHI ENDO** and HIROSHI ENOMOTO***

(1st Dept. Int. Med., Kyushu Univ.; *) Div. Cancer, Dept. Path., Nat. Inst. Health;

) TC-Lab., Inst. Infect. Dis., Univ. Tokyo; *) Dept. Physiol. Chem., Fac. Pharm. Sci., Univ. Tokyo.)

Two different cell strains were established in tissue culture from heart and liver tissues of Wistar rats, designated respectively as the strains JTC-4 and JTC-6. The two are considerably resembling to each other in the fibroblast-like appearance, growth rate and culture history until the establishment.

For the purpose of examining whether any difference or specificity would be found in the cell property between them which was characterized by that of the original tissue, the activity of collagen formation in tissue culture was compared in the present paper by the estimation of the content of hydroxyproline in the cultured cells.

At a time of initiation and in 2, 4, 7, 10 and 14 days of cultivation, the hydroxyproline content was determined following the method by Miyada and Tappel, 1956, simultaneously with the estimation of nucleus number in culture tubes.

During 14 days the cell population enlarged approximately 9.4 fold in the JTC-4 and 10.7 fold in the JTC-6. The amounts of hydroxyproline determined in the cultures were converted into those per cell unit. And distinct difference was demonstrated in the hydroxyproline content between the two strains. In the former the content developed approximately 2 fold during 14 days on the basis of per cell unit, while it remained almost unchanged in the latter.

As well as the findings by several workers in Japan, this result will indicate that some of the characteristics of the original tissue will be preserved among the cells even after a long time of cultivation in tissue culture.

This work was supported in part by the grants from the Ministry of Education, from the Ministry of Agriculture and Forestry, and from Asahi Shimbun Publishing Co.

235. FURTHER STUDIES ON "ONCOTREPHIN", THE MITOSIS PROMOTING SUBSTANCE IN MALIGNANT TISSUES

MASARU KURU, GORO KOSAKI, TAIJIRO MATSUSHIMA,
EITARO ITO, TAKASHI OGAWA and SHUN'ICHI NAKAMURA

(2nd Surgical Clinic, and the Institute for Cancer Research, Med. School, Osaka Univ.)

In the previous papers, the authors have reported that, oncotrephin, the mitosis promoting substance originally isolated from human seminomas and sarcomas, was present in fraction S_2 which precipitated from an extract of rat ascites hepatoma (AH 130) at 30-70 volume per cent of ethanol.

Fraction S_2 was heat stable, non-dialysable and by electrophoretic analysis, the effective substance was shown to be in a specific fraction which was characterized by its faint orange-yellow colour and by having a similar mobility to the β -globulin of human serum.

In the present study, the influence of proteolysis and acid-hydrolysis on its activity were investigated.

As a source of oncotrephin, a human hepatoma was used.

Inplace of fraction S_2 , 2 subfractions namely fraction S_2' (precipitating at 30-50 volume per cent of ethanol) and fraction S_2'' (precipitating at 50-70 volume per cent of ethanol) were prepared. Fraction S_2' was more effective than fraction S_2'' for the propagation of strain L cells. Acid-hydrolysis of fraction S_2' or treatment with pronase, abolished the stimulatory effect upon the propagation of strain L cells.

Trypsin treatment did not affect the activity of fraction S_2' .

The S_2 fraction, which was effective *in vitro*, also increased the mitotic rate in liver cells of normal adult rats 24 hrs. after a single intraperitoneal injection.

附 議

久留 勝：動物における発育の迅速な組織としては、悪性腫瘍のほか、Embryo および再生組織を挙げることができます。Embryo については早く Carrel が Trephon という物質の存在を提唱していますが、多数の学者の、多年の努力にかかわらず、その精製には成功していません。また再生組織としては再生肝を中心に、近年核分裂を促進する物質の探究が行われていますが、これまた生物学的方法で、その存在が確認された程度で、詳細については何も判っていないといつてよいと思います。私どもは悪性腫瘍から Oncotrephin を抽出しましたが、それと同じ抽出方法で、再生肝のみならず、鶏の Embryo からまた、L 株細胞の増殖を促進する物質をとり出すことに成功しました。これらの事実はある程度、増殖を促進する物質——これには Embryo、再生組織の中に存在するような正常なものと、腫瘍中に存在するような異常なものとを、区別することができると考えますが、それらの間の差異を明らかにし、究極においてこれらの物質の本態や、作用機転を明らかにする、道を開いたものと信じます。来春二月福岡のシンポジウムで、この方面の現在までの研究の結果の一斑をお話する機会があると考えます。

勝田：①もし生体内における腫瘍の正常細胞に対する作用を考えておられるのならば、正常細胞を維持状態（維持培地）において、それに添加した方がよいのではないか。

②また各種の細胞に対して平行してしらべる必要もあると思う。

③この仕事はこれまでにないと誰かが追試してみる必要がある。われわれも今後やってみたい。

神前：いろいろの細胞についての実験、とくに正常に近い組織培養細胞についてもやってみて見たいとは思いますが、とにかく現在の所、L株細胞に対する促進物質に焦点を合わせて、分離をはかっています。これはまた *in vivo* で肝細胞に対しても増殖促進的に作用するようです。できれば、この増殖促進活性を組織スライスについての生化学的な *mass stab* で検定したいと考えています。

佐藤春郎：本物質は *in vivo* で腫瘍の増殖を促進するというようなことはみられておりますか。

伊藤：*in vivo* で、発育の比較的遅い腫瘍に対する、本物質の効果はいまだ検討したことはありません。現在までには、正常肝臓に対する効果の検討を行っただけであります。

236. ISOLATION OF THE MITOSIS PROMOTING SUBSTANCE FROM REGENERATING RAT LIVER AND CHICK EMBRYO

MASARU KURU, GŌRO KOSAKI, YUKITOSHI AOKI,

SATORU MORISHITA, TAKEO UTSUNOMIYA and HIROSHIGE WATANABE

(2nd Surgical Clinic, and the Institute for Cancer Research, Med. School, Osaka Univ.)

In the previous reports, Kuru *et al.* have reported the existence of a mitosis promoting substance (or substances) in certain malignant tumors of man and named this substance oncotrephin. In the present paper the existence of such substances in similar rapidly growing nonmalignant tissues is discussed. As material for the work, regenerating rat liver and chick embryo were used.

A saline extract of regenerating liver of an adult rat, 48-72 hrs. after partial hepatectomy, was separated by ethanol into 3 fractions, i.e. the precipitate formed at 0-30 vol. % ethanol: fraction S_1 , the precipitate formed at 30-70 vol. % ethanol: fraction S_2 and the resulting supernatant: fraction S_3 . Fraction S_2 from regenerating rat liver like oncotrephin, promotes proliferation of strain L cells in simplified replicate tissue culture, while the other fractions were without effect. Fraction S_2 from normal adult rat liver or from regenerating liver which had been obtained within 24 hrs. or later than 120 hrs. after partial hepatectomy was not so effective. Effective fraction S_2 was inactivated by heating treatment at 100°C for 30 min., but not inactivated by similar treatment at 56°C for 30 min. The effective substance was non-dialysable.

The S_2 fraction, effective *in vitro*, also increased mitosis of rat liver cells 24 hrs. after a single intraperitoneal injection.

The stimulatory effect of a chick embryo extract and various fractions of it on

proliferation of strain L cells was also investigated. In this case too, fraction S₂ had much effect.

These results suggest that normal rapidly growing tissue also contains an oncot-rephin-like substance (or substances). A comparison of these 3 different mitosis promoting substances is now in progress.

237. TISSUE CULTURE OF MONKEY KIDNEY CELLS (I) ON THE NUTRITIONAL REQUIREMENTS

HAJIM KATSUTA*, TOSHIKO TAKAOKA* and ISAMU TAGAYA

(Tissue Culture Laboratory, Institute for Infections Diseases, Univ. of Tokyo.*;

Dept. of Virology and Rickettsiology, National Institute of Health, Tokyo)

Cortex cells from monkey kidneys were examined of their nutritional requirements in primary cultivation of tissue culture. Luxuriant proliferation was found in the medium consisting of bovine serum, lactalbumin hydrolysate and saline. The optimal concentration of the serum was 5%, but was 10% when dialyzed. The existence of serum proteins was essential to the proliferation and approximately 80% volume of these proteins were found to be replaced with the addition of 0.1% polyvinylpyrrolidone (PVP; AMW 700,000). By the use of PVP alone as the high molecular, considerable rate of proliferation was induced. The addition of chick embryo extract was apparently inhibitory on the proliferation under the concentrations which had been necessary for the propagation of embryonic cells, but was acceleratory in very low concentration. The optimal concentration of lactalbumin hydrolysate was 0.4% in both media with serum or PVP. Yeast extract was inhibitory under the concentrations commonly used. Continuous cultivation of these cells has been performed in a protein-free medium from their primary culture.

This work was supported in part by the grants from the Ministry of Education, from the Ministry of Agriculture and Forestry, and Asashi Shimbun Publishing Co.

**238. STUDIES ON STRAIN L CELLS (MOUSE FIBROBLASTS)
IN PROTEIN-FREE MEDIA. (VII) AMINO ACID RE-
QUIREMENTS IN SYNTHETIC MEDIA**

TOSHIKO TAKAOKA, HAJIM KATSUTA and KIMIE KANEKO

(Tissue Culture Lab., Inst. for Infect. Dis. Univ. of Tokyo)

From the strain L cells, four kinds of substrains were established in different protein-free media, designated as the substrains L_{P_1} , L_{P_2} , L_{P_3} and L_{P_4} . The media for the continuous cultivation of these cells consist respectively of 1) 0.1% polyvinylpyrrolidone (PVP; AMW 700,000), 0.4% lactalbumin hydrolysate (L), 0.08% yeast extract (Y) and saline (the mixture D); 2) 0.4% L and 0.08% Y in saline D; 3) the chemically defined synthetic medium, the mixture DM-12; and 4) 0.4% L in D.

The result in the sixth paper of minimum requirements of the L_{P_1} cells was re-examined again in this paper with special reference to the optimal concentration of each amino acid in the medium. And the amounts in the mixture DM-65 of Met, Try, Phe, CySH, Thr, Val and His were modified respectively to 8, 0, 10, 160, 200, 85 and 30 mg per liter in the mixture DM-114 which was obtained after the last examination.

Both of the mixtures DM-12 and DM-114 were compared of their nutritional activities for the four substrains. And considerable difference was demonstrated in the amino acid requirement among the four substrains. With the mixture DM-12 luxuriant cell proliferation was induced in all of the four, but not in all with the mixture DM-114 which was devised to be suitable to the proliferation of the L_{P_1} cells.

This work was supported in part by the grants from the Ministry of Education, from the Ministry of Agriculture and Forestry, and from Asahi Shimbun Publishing Co.

239. STUDIES ON THE ACTION OF HORMONES IN TISSUE
CULTURE (III) LEUCINE AMINOPEPTIDASE ACTIVITY
OF THE STRAIN HELA CELLS (CERVICAL CAR-
CINOMA OF HUMAN UTERUS) AND THE
EFFECTS OF SEX HORMONES

HIROYOSHI ENDO, KATSUMI WAKABAYASHI, HIROSHI ENOMOTO,
HAJIM KATSUTA* and TOSHIKO TAKAOKA*

(Dept. of Physiol. Chem., Fac. of Pharmac. Sciences, Univ. of Tokyo)
(Tissue Culture Lab., Inst. for Infect. Dis., Univ. of Tokyo*)

The proliferation of HeLa cells was shown in the 1st paper of the present authors to be stimulated by the addition of progesterone into the culture medium and inhibited remarkably by testosterone.

In this work the HeLa cells were examined of their leucine aminopeptidase activity, which had been histochemically demonstrated in the epithelial cells of normal human uterus. The activities were estimated by the colorimetric determination of β -naphthylamine splitted from the synthetic substrate, L-leucyl- β -naphthylamide, by the enzyme in the homogenate of the HeLa cells.

High activity of the enzyme was found in the HeLa cells and it was changed by the addition of sexual steroid hormones. Although no directed change of the enzyme activity was found in the earlier period of the cultivation owing to rather high degree of fluctuation, in the later period a stimulative tendency for the activity was evidently induced by the addition of testosterone. But progesterone and estradiol did not influence the activity.

The discrepancy was discussed between the effects of the hormones on the proliferation and on their leucine aminopeptidase activity of the HeLa cells.

This work was supported in part by the grants from the Ministry of Education and from the Ministry of Agriculture and Forestry.

附 議

山田文夫:「性ホルモン」を「子宮内膜細胞」である HeLa に作用させた場合直ちにホルモン剤が子宮細胞に作用をおよぼすとするよりむしろ一つの化学物質が一つの培養細胞に作用したと考える方がよろしくはないかとも考えるが、貴見承りたい。

遠藤: leucine aminopeptidase 活性に対する影響については、現在 HeLa 細胞しかやっておりませんが、細胞増殖の面では、鶏胚心線維芽細胞や AH-130 は HeLa 細胞のようなホルモンに対する感受性を示さない事実、また他の形態学的な研究で子宮の形質を *in vitro* に保持するには estrogen が必要である事実等々から、御報告致しました知見はやはり子宮粘膜細胞としての HeLa 細胞に黄体ホルモンとしての progesterone、卵胞ホルモンとしての estradiol、および男性ホルモンとしての testosterone が作用した結果と考えております。併しこれはあくまで一つの考え方でありまして、これが妥当なものか否かについてはさらに今後各種の非性器性の細胞に関する結果や *in vivo* の結果にてらして検討してみたいと考え

ております。

勝田：遠藤君の報告した他にわれわれは、HeLa に Testosterone を抑制濃度を与えておいてそれに女性ホルモンを各種濃度を与えたところ、抑制された増殖が女性ホルモン添加により回復され、しかも両者の間には一定の拮抗量比のあることを見出している。これはやはり、HeLa に対して一種のホルモン作用をおこなっていると考える方が合理的と思う。

240. STUDIES ON THE GROWTH FACTORS OF HELA CELLS BY DIALYZING CULTURE METHODS

SADAO KOZUKA (2nd Dept. of Path., Nagoya Univ.)

1. Single cell culture

Single cell culture technique is usually difficult in mammalian cells. In this study, cellulose tubing was employed to separate a single cell from untreated cell layer of 10^6 HeLa cells. Culture medium was a mixture of 20% bovine serum and 80% of 0.4% lactoalbumin hydrolysate-Earle's solution (L.E.). For 7 days culture, the mean generation time was 25.17 ± 1.04 hours in single cell lines with feeder layer, while a single cell without feeder layer was died or not multiplied. This result might indicate that medium was altered to more suitable one for cell multiplication by a large number of living cells; that is, small molecules which could easily pass through the dialytic membrane, participated on the alteration of medium, while large molecules such as serum protein did not concern at least directly.

2. To study on the role of serum protein on HeLa cells, dialyzing culture method was also employed. In outer side of cellulose tubing, 8×10^4 cells were cultured on the cover glass with 5 ml of the various media as shown on the table. Media in cellulose tubing of 2 ml in volume were also shown on the table.

Outer Part of Cellulose Tubing	Inner Part of Cellulose Tubing	Cell Growth
20% Serum+LE	20% Serum+LE	卅
DLE	40% DS	+
DLE	40% DS+ 10^6 Cells	+~卅
0.5% PVP+DLE	40% DS	+
0.5% PVP+DLE	0.5% PVP+DLE	±~-
0.5% PVP+LE	0.5% PVP+LE	-
DLE	DLE	±
LE	LE	-

DLE; Dialysate of serum in L.E. No protein was detected in it by sulfosalicylic acid.

PVP; Polyvinylpyrrolidone K-90

DS; Dialyzed serum protein

Forty-eight hours cultured cells were examined with H-E staining. Cell damage was prominent in L.E. alone, but slight in L.E. adding with low molecules of serum. High molecules, PVP K-90, was somewhat effective on maintenance of cell growth. Dialyzing serum protein kept in cellulose tubing was more suitable for cell growth.

This fact might mean that serum protein concerned not only to maintain osmotic pressure in medium, but also to supply low molecules wasted in medium. It may be conceivable that a detoxicative action occurs by combination of serum protein with toxic substances which existed already or produced by living cells.

附 議

勝田：① PVP を用いるなら，その至適濃度を用いなくてはならぬ。われわれの研究ではそれは HeLa では 0.1% であり，現にわれわれはそれを用いた無蛋白培地で 2 亜系を 1 年位継代培養している。また高分子を全く含まぬ完全培地で継代している亜株も持っている。貴説はその意味ですでに判っていることである。

② 対照として示された HeLa の形態は余りきれいに見えない。もう少し対照も健全に発育するようにしてから，実験することをおすすめする。

小塚：1) 無蛋白培地に移した初期の変化を示したのであり，PVP を用いた HeLa の継代株をつくられる初期には，やはりこのような強い変性が大部分の細胞に起ったときいています。私の研究の目的はこのような培養初期に変化の起らない無蛋白培地を目指しているのです。

2) 短時間の培養のため，移植の際，少数の細胞が変性を起したものが，そのまま残っているのは，むしろ当然と考えます。

241. CYTOPATHOGENIC EFFECT OF ANTISERA ON MAMMALIAN CELLS

KOICHI TAKANO and YASUKO HIROKAWA

(Dept. of Path., National Inst. of Health)

The colonial clones of HeLa cells, isolated from parent stock culture line and prolonged culture line by means of Puck's colony formation technique, were inoculated intravenously into rabbits repeatedly. The antisera thus obtained showed cytopathogenic effect (C.P.E.) not only on clones of HeLa strain but on other human cell lines as well, while they had no effect on L cells (derived from mouse) nor on JTC-6 cells (from rat liver). On the other hand, anti-L- or anti-JTC-6-serum showed C.P.E. only on homologous cell strain, and neither of them had any effect on HeLa strains. Clearly there is species specificity in C.P.E. of antisera on mammalian cells.

Morphological change of L and JTC-6 cells due to antisera were observed with phase contrast microscope. About 12 hours after the addition of antiserum in case of L cells and 7 hours in case of JTC-6 cells, the cytoplasm contracted to be dense, fine granules increased around the nuclei, though no change was detected in form

and interior structure of nuclei. After 20 hours in both cases, about 2/3 of the cells contracted into spherules. These contracted cells were deeply stained with Giemsa, thus showing features of pycnosis. Supravivally they did not take dye. The cells without conspicuous morphological change also had less ability to take dye than normal cells, which reveals the decrease of their physiological activity. There was no significant finding in PAS staining, methylgreen-pyronine staining and Sudan IV staining.

242. ELECTRONMICROSCOPIC STUDIES ON THE ROLE OF SERUM PROTEIN UPON HELA CELLS

SADAO KOZUKA and KIYOHIDE KOJIMA

(2nd Dept. of Pathol., School of Med. Nagoya Univ.; 2nd Dept. of Pathol., Med. School Nagoya City Univ.)

In order to clarify the role of serum protein on the HeLa strain, the cells that were cultured previously for 6 days in 5 ml of medium (0.4% lactoalbumine hydrolysate Earle's solution (L.E.) adding with 20% bovine serum), were investigated electron microscopically after 24 hours culture in the media as shown in table.

Medium	Degeneration of Mitochondria	Endoplasmic Reticulum	
		Elongate Form	Vesicular Form
A 20% Serum+80% L.E.	—~±	≡	±
B 0.5% PVP K-30 in L.E.	≡	+	≡
C 0.5% PVP K-90 in L.E.	≡	≡	≡
D Dialysate in L.E.	±	≡	+
E L.E. alone	≡	+	≡

PVP; Polyvinylpyrrolidone

Dialysate in L.E.; Dialysate from serum through cellulose tubing at 37°C for 24 hours in L.E. No Protein was detected in dialysate by sulfosalicylic acid.

The results were summarized in Table. In the B, C and E group, degenerative signs of mitochondria were noticed in the almost all the area of endoplasm, while in the A and D group, their degeneration was scarcely detected. Many of the mitochondria were swollen and the enveloping double membrane was disintegrated at several places. Irregular mitochondrial cristae, through vesiculation, broke up into granulations and disappeared. In some mitochondria, irregular cristae showed whirl formation occasionally, and converted themselves into osmiophilic substances. Further, osmiophilic

bodies interpreted as destroyed mitochondria were occasionally recognized. They were oval in shape and their limiting membrane was disintegrated. They were characterized by profile which was interpreted as deposition of osmiophilic substances into mitochondrial cristae.

Endoplasmic reticulums in a form of well elongated tubules were seen frequently in the A group, while in the B and E group, they tended to be disrupted to spherical vesicles with various size. In the C and D group, there were seen transitional forms between A and D or B group.

In Golgi complex, there could not be recognized any difference among each group. Glycogen areas were observed less in the B, C and E group than in the A and D group.

Based on these findings that submicroscopic changes of mitochondria were severe in the cells cultured in serum free media with or without addition of PVP, while their changes were scarcely seen in the cells cultured in the media with whole serum or its small molecules, it may be assumed that small molecules of serum play an important role on respiration of cells. Concerning to form of endoplasmic reticulum, it may be inferred that physical condition of medium plays one role in changes of endoplasmic reticulum.

243. INJURIOUS EFFECTS OF VISIBLE RAYS-IRRADIATION ON CULTURES OF HELA CELLS

KAZUO KAWAI and YOSHIO KATO (Dept. of Path., Mie Prefect. Univ.)

Visible rays (500-600 $m\mu$ wave-length) have been used for eye-observation or microcinematography of living cells by aid of Mohr's and Zettnow's adiabatic liquid filters. The present study deals with a problem, in what rate are culture cells affected by irradiation of these rays, using HeLa cells as a material.

An interphasic period of HeLa cells is about 30 hours by a method of perfusion chamber slide culture (Kawai & Isetani, 1959) and is about 15 hours by the cold shock method of parasynchronous division (Newton & Wildly, 1959). It was suggested that this difference is mainly due to the injurious effects of the visible rays; the parasynchronous growth curves and the intervals between the first and second bursts of division by the latter method are more effectively disturbed and elongated by frequently lighting for a short time than by lighting only once for the sum-total. Continuous irradiation with the weak light affects more intensively the cultures, especially the second burst of division than the first one. In addition, the cells in the dividing stages are more sensitive to the visible rays than in the resting stages.

Another appearance of the injurious effects is a marked reduction of cell adhesion

to glass immediately after lighting. Quantitative estimations of this property were made by the EDTA method (Kato, 1960).

244. THE EFFECT OF COLCHICINE ON THE HELA CELL

KIN'YA OKANO, KIYOKAZU NAGAI and YOICHI MORI

(2nd. Dept. of Path., Med. school, Osaka Univ.)

Formation of giant cells has been reported previously in cultivated HeLa strain cells that have been exposed to colchicine of various concentrations. In order to research more fully consequence of effected cells, observations were done.

HeLa strain cells were cultured constantly in the tube and its initial number was 10×10^4 cells per tube. Colchicine was added in various concentrations of 100 γ , 10 γ , 1 γ , 0.1 γ , 0.01 γ , 0.001 γ per cc on the 6th days of incubation, and allowed to react for 24 hrs. Thereafter, the cells were incubated in 80% lactalbuminhydrolysate and 20% calf serum solution. So, not only cytoplasmic division, but nuclear division were not performed in mitotic cells and its chromosome was scattered or formed clump. After 2 days, most of the effected cells left off the glass-surface, but the rest cells were survived and occassionally formed irregular polynuclear or mononuclear giant cells. On the statistical data, its distribution curve in the cell size broadened as the culture continued. Mean value of nucleolar number decreased after the effect, but reached the control value at re-proliferated stage. Mean value of the nucleo-cytoplasmic ratio was 0.250, but 5 days after 0.15 in 100 γ /cc, 0.16 in 1 γ /cc, and 0.231 in 0.01 γ /cc respectively. Most of the giant cells nucleo-cytoplasmic ratio were higher or lower than the control. Estimation of the nucleic acid was done according to the method of Schmidt-Thanhauser-Schneider, and at the 7th day, DNA contents of the HeLa cell was 38 γ /181 $\times 10^4$ cells (2.93×10^{-5} γ /cell), 29 γ /108 $\times 10^4$ (2.68×10^{-5} γ /cell) and 41 γ /144 $\times 10^4$ (2.84×10^{-5} γ /cell) effect, respectively 100 γ , 1 γ , 0.01 γ colchicine for 24 hrs. Otherwise, RNA contents of the cells for 6 days cultured was 64 γ /181 $\times 10^4$ (3.49×10^{-5} γ /cc), and 29 γ /108 $\times 10^4$ (3.38×10^{-5} γ /cell), 41 γ /144 $\times 10^4$ (3.54×10^{-5} γ /cell) effect respectively, 100 γ , 1 γ , 0.01 γ colchicine for 24 hrs.

The cell respiration was examined by the Warburg's manometric technique. In its stationary stage, logarithmic growth stage and colchicine effected stage oxygen uptakes were observed. Its values were 0.6×10^{-5} μ l/cell/hr, 1.0×10^{-5} μ l/cell/hr, 1.1×10^{-5} μ l/cell/hr, respectively 2, 6, 7 days cultured cells. When exposed to colchicine for 24 hrs. after 6 days culture, oxygen uptake was 0.6×10^{-5} μ l/cell/hr, 0.9×10^{-5} μ l/cell/hr, respectively for 1 γ /cc, 0.01 γ /cc colchicine.

XII. Metastasis

245. THE MOTILITY OF GASTRIC CANCER CELLS (CONTINUED)

JIRO FUJIWARA (2nd Dept. of Internal Med., Med. School, Osaka Univ.)

In the previous paper, the author reported that cancer cells from the surface of gastric tumor showed the motility in the thin and thick layer of coverslips as well as in the hanging-drop.

The motility of gastric cancer cells under various conditions is shown in this paper.

The same methods as previous report were applied. The number of moved and unmoved cells, the time, the distance, the duration and the rate of movement were examined as the indication of motility.

The results are as follows:

1. Obvious differences in the motility were not observed among different types of histology—adenocarcinoma, scirrhous, carcinoma simplex and gelatinosum.

2. When cancer cells showed the degenerations in various grades, the motility of these cells mostly deteriorated: in these cases the number of unmoved cells were markedly increased and most cells were stained with 0.05% eosin solution by Schreck's method. On the other hand very few cells were stained with eosin solution when the motility were active. As shown in the above, the motility of cancer cells depended on the living activity of cells, rather than the histological types.

3. No remarkable difference on motility of cancer cells was observed between the following two cases: 1) cancer cells added with sera from patient, and 2) cancer cells added with sera from the same blood type of healthy adults. The cancer cells also were not morphologically changed with serum from healthy adult. It is said that serum from healthy adult has anti-tumor activity, but above results are contrary to our expectation.

4. Instead of serum, if Hank's solution contained yeast and antibiotics were applied, the motility were remarkably activated. So, it would be presumed that the motility of gastric cancer cells *in vivo* is more active than that *in vitro*, because conditions are better for cancer cells.

(大阪対ガン協会研究費による)

**246. ÜBER DAS STUDIUM DER TRANSPLANTATION UND
METASTASE VON EXPERIMENTELLEN TUMOR VON
PLEURA, INSBESONDERE DIE INPLANTATION
ZUR VERSCHIEDENER ORGANE**

IWAO WATANABE, KIYOMI MORIKAWA, TOMISABURO KATSUI,
YUTAKA NAGAOKA, YUTAKA TAKAYANAGI and SHIN YAMAKAWA

(Ite Chirurgische Klinik von der Med. Univ. Osaka)

Es gelingt bereits, daß sich in unserem Laboratorium Sarkom in der Pleurahöhle weißer Maus einer Reihe von Na 2 entwickelt, indem mittels des 20-Metylcholanthrene in deren Innenraum durch verschiedenen Methode einschiebt. Ferner kann die Tatsache dadurch sichergestellt werden, daß kleines Sarkom-stückchen von einer Maus auf anderen subkutan übergetragen und auch die Metastase durch die Blutbahn in Lunge erzeugt werden kann.

Es gelingt auch die Generation dieser Ueberpflanzung jetzet bis zur XVIII Generation zu erreichen.

Wir können mittels von diesem Material die Ueberpflanzung zur verschiedenen Organe—in der Intra und Extradural-raum, ins Hals, in die Brusthöhle, in die Leber, in die Niere, ins Pankreas, in die Bauchhöhle und ins subkutane Gewebe von oben-
gesagten Mäuse—durchführen und sind auf die Entwicklung und die Matastase des Tumors und auf die Veränderung deren Pathologischen Anatomie zu untersuchen.

**247. STUDIES ON METASTASIS OF CANCER (VIII)
EXPERIMENTS ON LYMPH NODE METASTASIS**

HARUO SATO, YOSHIKO (KAWASHIMA) HIKICHI,
KUNIHISA HASHIMOTO and TOSHIO TAKAHASHI

(Cancer Research Lab., The Research Inst. for Tuberculosis and
Leprosy, Tohoku Univ.)

Lymphatic spread of cancer is one of the main routes of dissemination of tumor cells, and lymph node metastasis is quite common in human cancer, however, there are not so many adequate materials in experimental tumors to mimic the metastatic conditions into the lymph nodes.

The mouse ascites hepatoma (MH 134, MH 129F, & MH 129P) are the tumors which take 100% either in the ascitic or solid form when transplanted intraperitoneally,

intravenously, subcutaneously, or in the tail of the inbred C₃H mice and its F₁ hybrids. Among these three tumors, MH 134 possesses a very frequent, marked, particularly quick metastasizability into the lymph nodes.

When the tumor was inoculated in the subcutaneous lymph space of the tail, the frequency of lymph node metastasis was about 95%. The amputation of the tumor-bearing tail could hardly prevent the occurrence of metastasis and could hardly cure the hosts. The rate of tumor death with metastases was more than 80% in the mice, the tails of which were amputated 10 or 15 days after tumor inoculation, while that was about 70% in those of which tails were done 3 days after transplantation.

Based on these data, a screening program was advocated to select the effective substances which prevent the early metastasis of tumors into the lymph nodes. Thus for example, the amputation of the tumor-bearing tail and combined chemotherapy were carried out. In the present data, the effectiveness could be gained only in the earliest group of amputation (3 days after inoculation) when Mitomycin or Chromomycin was administered. The routes of the lymphatic spread in the tail were examined by means of a technique of direct lymphangiogram (mercury injection). The by-pass lymph vessels of the sciatic node were found on the way from the tail to the retroperitoneal (lumbar) nodes.

The materials used in this series of experiments are suitable for the basic studies of the mechanism in cancer metastasis.

(文部省科学研究費による)

248. STUDIES ON METASTASIS OF CANCER (IX) METASTASIZABILITY OF TRANSPLANTABLE MAMMARY CARCINOMAS IN C₃H MICE

TAKEO SAITO and HARUO SATO

(Dept. of Path., Fukushima Med. College; Cancer Research Laboratory, The
Research Inst. for Tuberculosis and Leprosy, Tohoku Univ.)

Several strains of mammary tumors which developed spontaneously in the C₃H inbred mice, have been maintained by means of serial subcutaneous transplantation.

The ascitic conversion of these tumors was unsuccessful except one tumor (reported in Gann, Vol. 50, Suppl., 138, 1959). The frequency of metastasis formation of this tumor (FM3A) was relatively high, but in other tumor strains, it was very low or almost none as seen in the table, where the general characteristics of these tumors such as transplantation rate, survival time and metastasizability were summarized.

When these tumors were inoculated subcutaneously, and the tumor nodules were extirpated surgically, there was no recurrence of tumor and the hosts were used to

be cured if the extirpation was complete and early enough.

We have been studying several transplantable tumors (mouse ascites hepatomas) which possessed a high and marked metastasizability (reported in Gann, Vol. 50, Suppl., 193, 1959). These tumors of mammary origin were also transplantable but not convertible into ascitic form. It seemed to be that the isolating ability and the growth speed of the tumors might be correlated with the metastasizing ability of a tumor.

Biological characteristics of the transplantable mammary carcinomas of C₃H mice

Tumor	Transplantation Rate	Survival Time	Metastasis	
			Primary Animal	Transplanted Animals
FM-2	93.7%	67.5(24~177)	—	1/32
3A	100.0	23.5(15~73)	}	(72%)
3S	98.55	63.4(21~178)		(25%)
4	100.0	137.7(34~303)		—
5	98.14	80.0(39~140)	+	—
6	100.0	67.1(45~118)	—	—
OM-6	85.7	99.08(34~301)	—	1/14
10	93.7	131.8(44~362)	—	2/32
14	98.85	81.7(22~221)	—	2/87
20	94.73	93.6(44~327)	+	1/38
23	93.6	57.3(28~118)	—	—
SM-1	91.3	59.0(27~154)	?	—

(文部省科学研究費による)

249. INTERCELLULAR ADHESIVENESS OF RAT ASCITES HEPATOMA CELLS AND METASTASIS

(II) INTERCELLULAR ADHESIVENESS AND NEGATIVE ELECTRIC SURFACE CHARGE IN "ISLANDS" OF ASCITES HEPATOMAS DURING ONE TRANSFER GENERATION

TAKASHI YAMADA (Dept. of Pathol., the Med. Inst. of Sasaki Found.)

In previous paper (Part 1) it was reported that the treatment of the rat ascites hepatoma islands with Tween solution *in vitro*, resulted to dissociate cells of the islands effectively. This "Tween effect" varied with the growth of various stage in each tumor strain.

In the present report this phenomenon was studied physicochemically, especially with reference to the mechanism of "Tween effect" as well as the relation between the effect and negative electric charge of hepatoma islands by colloid titration method (Terayama) during one transfer generation.

1. When hepatoma islands (AH 601) were agitated in the 1% Tween 80 solution at 37 C, the negative charge of the hepatoma islands was noted to increase temporarily during dissociations of cells, while the negative electric charge in the supernatant fluid increased with the laps of time (10-20 min.). The chemical analysis of the supernatant revealed that the depletion of cholesterol from the cellular surface of hepatoma islands was more marked. The depletion occurred earlier than that of nitrogen. This means that it was resulted from the early depletion of fat, comparing to the nitrogen, from the cell membrane through the dissociation of cells of hepatoma islands.

2. The negative electric charge of cell surface of hepatoma island (AH 601 and 149) varied as the proliferation of tumor cells during one transfer generation. This was parallel with "Tween effect" on the corresponding hepatoma island, without showing correlation to the size of hepatoma island. The electric negative charge in free-cell-type ascites hepatomas (AH 13, 66F, 62F and 414) increased also rather significantly in proportion to the cell proliferation. In other three tumors (AH 62, 602 and 7974), however, the negative electric charge was not so changed during their proliferation, showing the proportional correlation of "Tween effect" on them. The results suggested the mutual adhesiveness of hepatoma island cells varied with the proliferation of cells during one transfer generation, relating with the change of negative electric charge in the cellular surface.

250. AN EXPERIMENTAL STUDY ON METASTASIS FORMATION TO THE LIVER

KOICHI YOSHIDA, HISASHI WATANABE, KINGO YAMAGUCHI,
AKIO KOIZUMI and SUSUMU MAJIMA

(Prof. M. Muto's Surgical Clinic, Med. School, Tohoku Univ.)

Using ascitic hepatoma, such as AH 286, AH 7974, AH 66, AH 130, AH 66F, AH 13, and Y.S., injected into the extrahepatic portal vein of rats, we studied histologically immediate passage capability of tumor cells and formation mechanism of metastatic lesion in the liver.

1) Tumor cell thrombosis in peripheral branch of portal vein and sinusoid was observed in all cases of 32 rats sacrificed immediately after transplantation of tumor

cells and 51 rats sacrificed after 24 hours. Many of tumor cells showed degenerative changes 24 hours after transplantation. In total of 230 cases sacrificed on 3rd, 5th, 7th and 10th days after transplantation, 187 cases revealed proliferation of tumor cells in the liver in various grade, but 43 had no tumor cells.

2) Island type tumor cells such as AH 286 and AH 173 remained as minute foci of tumor cell accumulation in thrombosed peripheral portal veins and adjacent sinusoids until the 3rd day. After 5 days, however, they affected the adjacent interstitial tissue and became small nodular foci of metastasis surrounded by connective tissue. After 7 days these metastatic foci destroyed liver parenchyma severely and tumor cells poured into the portal vein system.

3) On the contrary, free cell type tumor cells like AH 66F, AH 13, Y.S. formed nodular foci less frequently. Mostly they grew diffusely in the sinusoid and drained into the central vein or hepatic vein in early stadium. In addition, mixed type tumor cells like AH 7974, AH 130 formed both nodular and diffuse metastatic foci.

3) In order to study the immediate passage capability of tumor cells in the liver, blood was sampled from inferior vena cava just proximal to the liver at the same time of transplantation and tumor cells were examined in 32 rats sacrificed immediately after transplantation. None of 8 cases of island type tumor cells was positive, while 15 of 24 cases of free and mixed type tumor cells were positive, showing immediate passage capability through the liver.

251. THE GROWTH OF DIFFERENT TUMOR STRAIN IN DIFFERENT ORGANS FOLLOWING THE TRANSPLANTATION INTO THE LEFT CHAMBER OF THE HEART

TAKASHI YAMADA, LEONARDO ADACHI* and HIDEHIKO ISAKA

(The Med. Inst. of Sasaki Found.)

(Hospital Central del Empleado, Lima, PERU*)

Comparative studies of various strains of the ascites hepatoma in the rat were performed with special respects to the organs in which the tumor of each strain developed, after the injection of tumor cells into the left heart chamber of rats, and the frequency of its growth. The tumors used were every 4 kinds of free-cell-type tumor, AH 13, 414, 62F and 66F, and of island-type tumor, AH 66, 62, 601 and 7974. Almost all constituents of the former 4 tumors are individually isolated hepatoma cells, while the latter 4 are composed predominantly of clusters of hepatoma cells in the tumor ascites.

The tumor growth in the distant organs from the heart was noted in 57 among

the 174 rats which were injected effectively with tumor cells and survived long enough for the observation of tumor development. The results were as follows:

1. The rate of tumor growth after the intracardiac injection of the free-cell-type tumors was less than 50%.
2. The growth of the free-cell-type tumors was found most frequently in the kidneys, but seldom in the brain and the spleen. In general, the island-type tumors showed few growths.
3. Differences were noted in the organs in which the tumors developed frequently.

Organs	Developed tumors	
	frequently	rarely
Lungs	AH 13	
Ovaries	AH 414	AH 66F
Muscle	AH 66F, AH 414	AH 62F
Eyes	AH 7974	

4. The results were discussed from the viewpoint of blood-borne metastasis of various tumors derived from the common normal ancestry, the liver cell. It was suggested that the above difference in the metastatic growth might be due to different biological characteristics of each tumor.

This investigation was supported by Grant CY-2799 from the National Cancer Institute, NIH, U.S. Public Health Service.

252. STUDY ON METASTASIS OF GASTRIC CARCINOMA (A CLINICOPATHOLOGIC CLASSIFICATION OF GASTRIC CARCINOMA)

SHIRO KITAMURA, KUNII KOJIMA, KOKICHI TSUCHIYA
and SHOICHI WATANABE

(Dept. of Path. and Dept. of Orthop. Surgery, Fukushima Med. College)

The authors investigated the relation between the primary gastric lesion and metastasis in 48 autopsy cases of gastric carcinoma. The results revealed a close relation between them. Gastric carcinoma was observed from the whole living body and classified into the following three types clinicopathologically: The first type was adenocarcinoma, the second medullary and simple carcinoma, the third scirrhous and mucoid carcinoma.

Adenocarcinoma was mainly localized at the pylorus or the corpus and formed a

large tumor. In metastasis to perigastric lymph nodes, the metastatic lymph nodes were large and nodular and had a conglomerating tendency, existing mostly near the lesser curvature and the pylorus. Furthermore, in metastasis to more remote lymph nodes, organs and tissues, adenocarcinoma metastasized dominantly to retroperitoneal and hepatic lymph nodes, the liver and pancreas, and those metastases also were large and nodular. However, the metastases were rare in mediastinal lymph nodes and more remote organs such as the lungs, bone marrow and genitourinary organs. As to the peripheral blood picture in adenocarcinoma, only shift to the left was observed in most cases.

On the other hand, the primary lesion of scirrhus and mucoid carcinoma usually infiltrated almost into the whole stomach diffusely and the metastatic lymph nodes were small and swollen solitarily, and observed evenly in any perigastric region. The rate of metastasis of scirrhus and mucoid carcinoma to the liver and pancreas was lower than that of adenocarcinoma, but this type of carcinoma often metastasized to the lungs, bone marrow and genitourinary organs in form of carcinosis. The metastases in the intestinal tract and peritoneum were widespread and those in lymph nodes were evenly disseminated. In the peripheral blood picture, leucemoid reaction with myeloblasts was increasingly seen in this type.

Simple and medullary carcinoma had an intermediate behavior between the first type and the third on the primary gastric lesion, metastatic state and also the peripheral blood picture.

From the other viewpoint, this classification was based on the degree of mutual disjunction of carcinomatous cells.

附 議

岡野錦弥：先年教室の桐本が胃癌の原発巣と転移巣の組織学的比較を大阪大学医学雑誌に発表したのが、この際 Kaufmann の分類によると原発巣の全組織が単一の組織学的表現をなし得るものは約 10% 前後であった。貴研究の組織学的分類の際の組織像の混合に対する御意見をお伺いします。

今井 環：われわれの所で見たところでは、同じくびまん型癌（北村教授のいう硬癌）に、リンパ節転移型と腹膜播種型があるが、前者には、原発部にリンパ管内蔓延型（L 型）の像の多少とも著明なことが多い。

肝転移型では、腺癌型を主とするものが多く、一般に高年に傾いている。

びまん型癌の肝転移頻度に関する北村、村上両教授の所見の差は、剖検例と手術例の差と関係あるかも知れないと思う。われわれの剖検例でも、北村教授の示すような所見を得ている。

武田：胃癌の分化と転移との関係はきわめて興味がある、このさい癌の分化の外に発生年齢が転移の頻度および方向に関係はないか。

村上忠重：手術した後の胃癌患者の解剖所見では腺癌の肝への転移の多いのにくらべて硬癌はきわめて少ない。しかるに演者の成績ではこれが少くない。これを血行性のものと考えられるか蔓延性のものと考えられるか教示されたい。

田内 久：年齢の差と胃癌組織像の差については私どもも以前から注目していますが、始めは、年齢の差による発生母組織の差も問題にしたこともあるが私どもの最近の検索によると、比較的早期と考える手術例では年齢の差による組織像の差がいちじるしくないにもかかわらず、比較的晚期と考えられる剖検例では年齢差による組織像の差が顕著であるような成績を得ています。（もちろん年齢差による組織像の差の相違は演者ならびに武田教授の御話と同様です。）このような事実は癌の発育過程における癌組織像の変換の

様相が個体の年令差によって種々の修飾をうけることを物語ると考えられる。私どもはこの場合年令差による間質の増生態度の差の影響も重視した。

滝沢延次郎：私は癌の転移のあるものと転移のないもので原発巣に組織学的に差があるように思っておりますが演者の例の第1型の中に転移のなかった例はありませんでしたか。

演者：1) 岡野君へ。時間の関係で説明いたしませんでしたが、スライドに示しましたように、原発巣は部位によって種々の組織像を呈します。そのうちの主組織像をもって組織学的に分類しました。概括的に見て、腺癌は大体単純癌くらいまで移行し、殊に小腺腔のものは硬癌の像を一部に示すこともあります。逆に硬癌、膠様癌、単純癌は多くは互にその範囲内で移行を示し、ときには小腺腔を有する腺癌に移行します。

2) 今井君へ

硬癌にリンパ行性転移を主とするものと腹膜転移を主とするものがあるとのことでありますが、この点については材料を再検討して見ます。

3) 武田君へ

年令的に見ますと40才以上の人に腺癌が多く、40才以下の人に硬癌が多くなっています。腺癌は幽門、体部に限局いたしますし、硬癌はびまん性に拡がります。

4) 村上君へ

硬癌の際の肝転移は肉眼的に見られることは少く、組織学的に証明されるもので、それも肝の横隔面では少く、肝門の近くの「グ」氏鞘内に *Carcinosis* の形で浸潤しています。この *Carcinosis* の起し方にはもちろんリンパ行性のものもありますが、*Lungen Carcinose* の問題もありますので、リンパ行性か血行性かをさらに検討させていただきます。

5) 滝沢君へ

非手術の剖検例で全然転移のないものはありませんでした。ただ硬癌の1例で、胃周囲リンパ節がただ1個豌豆大に膨脹しただけのものがあります。このものは *Meningeale Carcinose* を起しておりました。概して胃周囲の局所リンパ節の転移が小さいもの程広汎な転移を示すように思われます。その点炎症に際して初感染群の反応の強いもの程全身への蔓延が少く、初感染群の反応の弱いものほど蔓延が強いことと同様のことが癌にもいえると思います。

253. A STUDY OF THE MECHANISM OF CANCER METASTASIS

SHUHEI TAKITA, HAYAMI NISHIJIMA, YASUO KISHINO,
SHIGERU TOYOTA and TAKASHI KAWAHIGASHI
(1st Dept. of Surgery, School of Med., Tokushima Univ.)

Our clinical cytological observation has confirmed that tumor cells in the ascites obtained during abdominal operations were detected in 19 out of 26 cases. In the peripheral blood the cells were found positive in 7 out of 18 cases, indicating possibility of hematogenous metastasis. In rabbits and rats, intraperitoneal spreading of the experimentally implanted tumor cells (i.e. Yoshida and Brown-Pearce tumor-cells) was found to be greatly modified by the presence of peritoneal adhesions.

Intravenous injection of Yoshida tumor-cells resulted in the development of liver metastasis in the region injured by CCl_4 or allyl formate, where seromuscular layers had been incised and sutured. Implantation of Yoshida tumor cells into the walls

of appendix was followed by appearance of the tumor cells in the peripheral blood. The implanted tumor was removed subsequently. It was concluded that the survival rate was the highest in the group which received Trespamin prior to the implantation of the tumor cells.

附 議

田崎勇三：われわれも数年前から Fibrinogen 法によって流血中の癌細胞の研究をしているが、貴兄の御発表と同様に胃癌根治手術後 3 日、6 日、10 日間において肘静脈血中に 10%, 33%, 40% の割合に癌細胞を証明した。何故に 3 日間より 10 日間へと増加するかについては目下研究中であり、10 日間以後の末梢流血中の癌細胞についてはいまだ検索を加えていない。

流血中の癌細胞については、いろいろの意見もあるが、われわれは太田教授の御援助によってこの研究をつづけている。しかし決定困難の細胞は疑陽性細胞として、真に癌細胞と考えられるものから除外しているが、それでも上述のごとき成績を得ている。

これらの流血中の癌細胞がすべて将来転移または再発の原因になるというのではないが、その中のある強力な細胞はどこかに着床して将来発癌のもととなり得る可能性は十分あると考えられるのである。しかもこれらは根治手術可能な比較的早期と思われるものであるから、胃癌の手術後には化学療法を併用したがいと考える次第である。

桂 重次：ただいま田崎会長の発言のごとく胃癌根治手術後 10 日で最も多く、癌細胞の血中出現があることが事実とすればこの癌細胞出現の意義が種々問題を含んでいると思われる。今後十分検討してみたい。

254. EXPERIMENTAL STUDIES ON THE CONDITION OF THE INTRAMURAL COMMUNICATION OF THE TISSUE FLUID IN THE GASTRO-DUODENAL WALL

KATSUJI HAYASHI, SEIICHI HIROSE, SHIGERU OGAWA
and HISAMASA SATO

(1st Dept. of Path., Med. School, Nagoya City Univ.)

In order to study the mechanism of invasion of gastric cancer into the duodenal wall, we investigated the relationship between the motility of the gastric wall and the communication of tissue fluid of the wall of the two viscera using india-ink.

In a preliminary investigation in which rabbits were used, it was clarified by X-ray examination that the application of vagostigmin, besacolin or atropin was effective to change gastric motility.

When albino-rats received atropin, the injected india-ink in the gastric wall easily communicated into the duodenal wall and on the contrary in rats which received vagostigmin or besacolin, it moved with difficulty.

The above findings may suggest that lowered peristalsis of the stomach may accelerate the communication of the tissue fluid in the gastric wall into the duodenum.

255. STUDIES ON THE SPREAD OF THE RECTAL CANCER BY MEANS OF RESECTED SPECIMENS

HIKO IKEUCHI and SHIGERU OGAWA*

(2nd Dept. of Surgery & 1st Dept. of Path.,* Med. School, Nagoya City Univ.)

Abdominoperineal combination method with colostomy (Miles) is the most common operation for rectal cancer. As compared with this, Babcock-Bacon method is more excellent when the indication is correct. To determine the radicality by the latter, cancerous invasion was histologically investigated on the sections from 54 specimens surgically removed.

Regarding to cancerous spread, tendency to penetrate the intestinal wall in the surrounding tissues was more marked comparing to superficial spread.

As to the route of spread in those specimens, it was almost lymphogenous and less hematogenous.

Further, rectal cancer was more common in 40 to 60 years old with no difference by sex, and they showed ulceration about 4×4 cm. in size macroscopically in this series.

These findings obtained will suggest that Babcock-Bacon method should be indicated only in the case, with restricted metastasis, in which lower end of tumor was observed in the site fairly remote from anorectal line. Provided that the application is accurate, radicality by its operation might be quite sufficient.

附 議

北出文男：口側ならびに肛門側にそれぞれ 2~3 cm の浸潤を示した例において、浸潤の存在部位は粘膜下組織でしたでしょうかあるいは筋層より外側に存在したでしょうか。

池内：肛門側 3 cm, 口側 2 cm におよぶ浸潤の直腸壁における分布は全て筋層あるいは筋層以上において浸潤していて粘膜、粘膜下層には癌性浸潤はみられなかった。

今井 環：術後遠隔成績との関係をお調べになっておれば、御教示いただきたい。

池内：予後の関係は本手術症例 (Babcock-Bacon) が少数のため今後症例を追加検討する予定である。

256. PATHO-HISTOLOGICAL STUDY ON THE RELATIONSHIP
BETWEEN PRIMARY LESIONS AND LYMPHNODE METAS-
TASIS IN 163 CASES OF THE OPERATED PATIENTS
WITH UTERINE CERVICAL CANCER

TAKASHI KOBAYASHI, SHOSHICHI TAKEUCHI,

KAZUO MATSUEDA and KOOJI YAMADA

(Dept. of Obst. & Gynec., School of Med., Univ. of Tokyo)

One of the most important criteria in determining the prognosis of uterine cervical cancer at present, is lymphnode metastasis. Various pathological findings relating to metastasis of lymphnodes have been reported. These include factors pertaining to size, degree of cell differentiation (Mertzloff and Broder), nuclear changes (Black), stromal changes of the primary lesion, nuclear changes (lymph-vessel permeation type, or so-called CPL classification) and sinus histiocytosis of the uninvolved lymphnodes (Black). All of these factors were evaluated from the pathological point of view for indications of lymphnode metastasis.

Material: Radical panhysterectomy with extended lymphadenectomy was performed on 163 cancer cases and 2060 lymphnodes were obtained. These cancer cases consisted of 45 cases in the first stage, 102 cases in the second stage and 16 in the third stage of cancer. The incidence of metastasis was found to be 17.8%, 27.5% and 62.5% in the respective stages.

Results: 1) The most significant factor relating to metastasis was found to be the depth of infiltration in the primary lesion. ($\alpha < 0.01\%$). 2) Lymph vessel permeation or Imai's CPL classification was also significant to determine the presence of lymphnode metastasis but not as well. ($\alpha < 10\%$). 3) Broder's classification did not coincide with the presence of metastasis in all cases, but significant relation between them was found, when the depth of the primary lesions were analyzed and taken into consideration. ($\alpha < 5\%$). 4) No significant correlation was found to exist between other factors such as Mertzloff's classification, nuclear changes (Black), classification advised by the Japanese Uterine Cancer Committee, sinus histiocytosis of the uninvolved lymphnodes (Black) and metastasis of the lymphnodes, even when the depth of the primary tumors were taken into consideration.

(文部省科学研究費, 厚生省研究費による)

附 議

藤森達水: 淋巴節の転移を調べるのにどういう方法によりましたか, またそれについて何等かの科学的根拠がありますか。

私の所では推計学的理論に基づいて, 検出率と転移率とを計算しています。この内容は次の 257 番で発表します。

松枝：われわれのリンパ節の検索方法は剔出リンパ節をできるだけ多数についてそれぞれの中央部切片を一枝宛作成し鏡検する方法です。ときに脂肪組織のみよりなるごとく見えることがあります、その際は脂肪組織ごと包埋します。

転移のあるリンパ節は必ずしも腫大せず極めて小さく米粒大のことすらあり、逆に腫大しても単なるリンパ細胞増生によるか炎症による髄様腫脹で転移のないことがあり、リンパ節の大きさと転移との関係は一定しない。また、リンパ節転移には一定の径路順位が考えられるわけであるが、子宮頸癌の場合、一次リンパ節というべきものに数種をあげることができる。

したがって特定少数の腫大リンパ節材料についての検索のみで、その症例の転移の有無を云々したり、転移率を算定したりするのは危険が多くなるのではないかと考えております。

滝沢：演者の例で転移のない子宮癌と転移のある例との間に組織学的に癌実質組織に差違はありませんでしょうか。

松枝：われわれの成績でも転移のない場合の原発巣の組織像は比較的分化度の高いものが多かった。すなわち同一の浸潤の深さを示す原発巣で相比较で見ますと Broder 分類が推計学的に有意でした。

今井 環：C.P.L. の場合、当該腫瘍が C とか P とか L の何れかというだけでなく、各型の grade を取入れていただいたらと思っている。I 度と III 度とでは、同じ L でも、相当違うのではないかと感じるわけである。

松枝：C.P.L. それぞれの程度をさらに分類して、観察を行っていますが、現在まだそれぞれの例数が少ないので、今回は御質問の点についての統計的観察を省略させていただきました。例数の増加をまって後報の予定でおります。

257. A STATISTICAL STUDY ON PELVIC LYMPHNODE METASTASES OF CARCINOMA OF THE CERVIX

HAYAMI FUJIMORI and YASUSHI KASAHARA
(Dept. of Obst. and Gyne. School of Med., Osaka City Univ.)

Forty-nine specimens from radically hysterectomized patients by Okabayashi's operation, who were diagnosed as carcinoma of the cervix (19 cases of clinical stage I, 26 cases of stage II and 4 cases of stage III) in our department, were macroscopically examined, so that the largest lymphnode of the right hypogastric chain was removed per each specimen. Two different methods were employed for the examination of each node microscopically.

After the multiple sections were taken from each specimen, firstly 3 slides were made by Sugiyama's statistical theory for each node. Secondly, only one slide was selected from each node after a random sampling.

According to the former method, the metastasis was found in 28.26% of all patients of stage I, and 30.91% in stage II. There is no significant difference statistically between these two groups. The latter investigation, however, revealed that if only one section was made on each of the specimens, the liability of such a section is 44.8% to be positive for the metastasis for the patients of stage I, and 68.7% for the patients of stage II. The difference between these two is significant statistically.

附 議

小林 隆：ただいまの発表と関連があるかと思うので、追加的な発言をしたいと思います。すでに発表しましたようにわれわれはリンパ節廓清効果を求めるのには、従来のようにリンパ節転位陽性例のみの治癒率で計算するのは不十分で、さらに潜伏転位例を理論的に計算し、これをも加えたものすなわち陽性および潜伏の転位合計例について治癒率を計算し、これと実際の治癒率とを比較して、外科的廓清の Criteria とした。

田村：実測度数が非常に少い組 (0 とか 1 とか) をもつ標本に χ^2 検定法を用いてよいか。

笠原： χ^2 の利用の疑問

現在の症例数が少ないためその結果に影響が大ということのようですがさらに検討を加えた上返答をいたしたいと思います。

258. STUDIES ON HEMATOGENIC METASTASIS OF CANCER

NOBUJIRO TAKIZAWA and KIKUO NAKANO

(Dept. of Path., School of Med., Chiba Univ.)

It was reported by us at the 3rd Cancer Symposium in 1960, that fifty-five autopsy-cases of carcinomas of various organs were classified into three grades on the viewpoint of lymphogenous metastasis. The result presented in that report were summarized as follows. In the three cases which were classified into the 1st grade no metastasis to the regional lymphnodes was a remarkable feature. Moreover, the histologic characteristics of the primary foci of carcinomas were highly differentiated. The seven cases of the 2nd grade A showed metastasis to the regional lymphnodes only microscopically, and the primary tumors of the carcinomas were those with moderated differentiation. The 2nd grade B consisted of five cases had regional lymphnode metastasis which were recognizable even macroscopically. In these cases the histologic appearances of the primary carcinomas were those of poor differentiation. In the 3rd grade we had fourty cases which were characterized both by metastasis to as well as beyond the regional lymphnodes and undifferentiated primary carcinomas. It is the purpose of this presentation to report the studies on the hematogenous metastasis especially to the lungs and liver of the same materials as reported at the above mentioned 3rd Cancer Symposium in 1960.

(1) All 3 cases of the 1st grade had no metastasis to the lungs and liver. One case of the 2nd grade A demonstrated a ricecorn sized metastasis to the liver, while a case of the 2nd grade B showed some cell-accumulations which were suspicious of metastatic carcinomas in the lungs. Thirty-six cases of the 3rd grade had metastasis to the lungs, liver or both. Therefore, it appears that the pulmonary and hepatic metastasis of the carcinomas is roughly proportional to the extent of the lymphnodes metastasis.

(2) In the 1st grade no carcinoma cell emboli were noted in the blood vessels of the primary foci, while the primary foci of the carcinomas in the 2nd and 3rd grades had more or less blood vessel invasions by tumor cells. Such carcinoma cells were more undifferentiated in the cases of the 3rd group than in the 2nd group.

(3) When carcinoma cells of early metastasis to the alveoli or hepatic lobules demonstrated active growth, the lining cells of the alveolar walls or stellate cells adjacent to the tumor cells were generally degeneratively atrophic, while they were well preserved when metastatic carcinomas were degenerative or disintegrating.

259. CYTOLOGICAL CRITERIA OF CANCER CELLS IN THE CIRCULATING BLOOD

HIDEO MUNAKATA, TOMOYUKI OKAWA and MASASHI OSAWA

(Dept. of Path., Dept. of Obst. & Gyne., Fukushima Med. College)

It seems to be the most basic necessity to have accurate criteria for identification of cancer cells in the circulating blood. In this study, the sizes and the areas of cell body, nucleus, and nucleolus, were measured, and average diameter, nucleo-cytoplasmic ratio, and nucleus-nucleolar ratio were calculated. As the materials, three kinds of samples were employed; smears of the blood, imprints and frozen sections of the tumor tissues.

The samples examined were obtained from the autopsied or surgically operated patients, among which cancer cells were detected in 15 malignant tumors, including 6 of chorion epithelioma malignum (CHR), 4 of stomach cancer, and others.

1. The average of the longest diameter of cancer cells in the blood, was about 27μ in those of CHR, and about 20μ in other cancers, respectively.

2. The maximal diameter was 73.8μ in the cells of CHR, and 36.6μ in other cancers. The minimal diameter was 13μ in the former, and 8.5μ in the latter.

3. The average size of tumor cells found in the smear preparations of the blood was bigger than that of tumor cells obtained in the imprints of frozen sections of the tumors, in general.

4. The nucleo-cytoplasmic ratio was 0.34 in CHR, and 0.49 in other cancers. The nucleus-nucleolar ratio was 0.048 in CHR, and 0.026 in other cancers. These indicate the characteristic features of tumor cells of the CHR.

5. Clumps of tumor cells were sometimes observed in the samples obtained from the right heart blood or the blood of the draining vein. The largest clumps contained more than 100 cells, and the largest diameter was more than 480μ .

260. CLINICAL SIGNIFICANCE OF TUMOR CELLS IN THE BLOOD STREAM IN PATIENT WITH GASTRIC CARCINOMA

SUSUMU MAJIMA, HISASHI WATANABE, KINGO YAMAGUCHI,
AKIO KOIZUMI and KOICHI YOSHIDA

(Prof. M. Muto's Surgical Clinic, Med. School, Tohoku Univ.)

Of 160 patients with cancer of the stomach who underwent surgery in the authors' clinic, tumor cells in the peripheral circulating blood was examined preoperatively. At surgery, sampled blood, from peripheral vein, draining vein from the lesion, and from the extrahepatic portal vein were examined. Results obtained were as follows:

1) In the peripheral blood, 7 of 160 cases (4%), in the local blood, 41 of 160 cases (26%) and in the portal blood 8 of 50 cases (16%) were positive for cancer cells.

2) Comparing the frequency of appearance of cancer cells into the blood stream in the curable and palliative cases, the positive cases for cancer cells were present in 25 of 118 curable cases (21.2%), and 17 of 42 palliative cases. (40.5%)

3) In regard to relation between the histological findings of gastric cancer and the frequency of tumor cell appearance in the blood, following results were obtained:

a) According to the classification of peritonitis carcinomatosa (Muto), 2 of 9 P.C.-O cases, 13 of 65 P.C.-1 cases, and 27 of 76 P.C.-2 to 3 cases were positive for cancer cells respectively.

b) In relation between lymph nodal involvement and cancer cells in the blood, 3 of 30 cases without lymph nodal involvement, 6 of 27 cases with primary lymph nodal involvement, and 33 of 103 cases with secondary lymph nodal involvement were positive for cancer cells respectively.

3) In relation to the grade of malignancy by Broders, 6 of 39 cases of Broder I to II, and 31 of 99 cases of Broder III-IV were positive for cancer cells.

261. CANCER CELLS IN CIRCULATING BLOOD OF THE PATIENTS WITH CANCER OF THE ESOPHAGUS (II)

MORIMITSU OHNISHI, FUMINORI YANAGISAWA, TADAO KAMATA,
ISAO MATSUZAKI, MASANAO KAWANA and TETSU YOKOYAMA

(Nakayama Surgical Dept., School of Med., Chiba Univ.)

The investigation for cancer cells in circulating blood was carried out on 27 patients with cancer of the esophagus. Blood samples were obtained by antecubital

vein puncture and cancer cells were isolated by the silicone method from whole blood. The identification of cancer cells was confirmed with its corresponding cytologic observation of smear taken from the primary lesion.

Of the 27 patients with cancer of the esophagus, 6 (8.3%) were positive and 12 (16.7%) were suspicious. The patients were further divided into curable group that were treated by a curative type of surgical procedure and incurable group. Two (6.1%) of the 33 curable patients were positive, and four (10.3%) of the 39 incurable patients were positive. No lymph node metastasis was found in the 15 patients who were treated by a curative type of surgical procedure. In this group, cancer cells were not found in circulating blood taken by antecubital vein puncture. In contrast, 2 (11.1%) of the 18 patients who had lymph node metastases were positive.

Twenty patients with cancer of the esophagus had a radiation therapy preoperatively and two positive patients who were irradiated more than 2,000 r. tumour doses changed into negative after the irradiation.

Cancer cells in circulating blood during an operative procedure were searched on 14 patients. In 3 (21.4%) of this patients, cancer cells were not found preoperatively but were found during the procedure in the peripheral circulating blood.

262. CANCER CELLS IN THE BLOOD OF LUNG CANCER PATIENTS (IV)

KAZUO WAKASA, SATOSHI KONDO and KUNIHISA HASHIMOTO

(The Research Inst. for Tuberculosis and Leprosy, Tohoku Univ.)

Cytological investigation of the cancer cells in the circulating blood, by means of the original Sandberg method, was carried out in 64 cases with primary lung cancer.

(1) Cancer cells were detected in 24 cases (37.5%) out of 64 subjects in the peripheral venous blood. In 9 of 14 cases examined at the time of operation cancer cells were also detected in the blood of the tumor draining vein.

(2) The grade of appearance of the cancer cells was higher in the blood of the tumor draining vein than that of the peripheral vein.

(3) The detection rates of the cancer cells in various stages of lung cancer were 20% in the silent stage, 33.3% in the urgent stage and 66.7% in the rampant stage; higher rate in the last stage than in the earlier stage.

(4) From the view point of surgery, the numbers of the detection positive cases were 7 in the 22 operated subjects, 3 in the 8 exploratory thoracotomy cases, 13 in the 29 inoperable cases and 1 in the 5 cases which were not surgically treated; higher detection rate in the inoperable and exploratory thoracotomy cases than in

the operated cases.

(5) Of the 7 detection positive cases, repeated examinations were made before, during and after chemotherapy. In 4 cases of these subjects, cancer cells were no longer detected after chemotherapy and in 1 case degenerated cells appeared during the course of chemotherapy. It is considered that chemotherapy may give an effect on the features and the fate of the cancer cells in the circulating blood.

263. A SEARCH FOR CANCER CELLS IN THE CIRCULATING BLOOD OF THE PATIENTS HAVING GYNECOLOGIC CANCER, WITH SPECIAL REFERENCE TO OPERATIVE PROCEDURE

TAKASHI KOBAYASHI, SHOSHICHI TAKEUCHI, TATSUHIRO KASAMATSU,
HIROSHI KOIZUMI, KAZUO MATSUEDA, TAKEHIKO YOSHIDA,
KOJI YAMDA, MAKOTO MURAKAMI and TSUYOSHI SUGIMOTO

(Dept. of Obst. & Gynec., School of Med., Univ. of Tokyo)

In the previous report "5 point marking classification" was advised as criteria of the atypical cells in the blood, because of difficulties encountered in making differentiation between malignant cells and abnormal cells which appeared into the blood apparently due to erythro-leucemoid reaction of the patients with cancer.

In this communication we wish to present preliminary data concerning our findings to date, with special reference to surgical manipulation.

Materials and Method; Before operation five ml of the blood were drawn from the antecubital vein as peripheral blood. During operation the same doses of the blood were also drawn for three times from the common iliac vein as regional blood; after extirpation of the pelvic lymphnodes, after resection of the cardinal ligament, and after extirpation of the uterus. The blood specimens were prepared with the method of silicone flotation (Seal), and the sedimentation method using acasia-glucose (Modification by Munekata). The atypical cells found in the blood specimens were marked into 5 point, based upon the previously reported criteria.

Results; 1) Preliminary results on 35 cases of gynecologic cancer patients were shown in Table 1. If only 5 point cells could be taken for cancer, the cancer cells were presented in only 1 case (2.9%), and if the atypical cells above 4 point could be interpreted as cancer, the cancer cells were found in 10 cases (28.6%).

2) Relationship between regional blood and peripheral blood: In 9 patients with cervical cancer, the atypical cells above 4 point in regional blood were presented in 2 cases (22.2%), while the cells above 4 point in peripheral blood were presented in 1 case (11.1%).

Table 1

No. of cases	Mark	No. of positive cases	per cent
35	above 3 point	15	42.8
	above 4 point	10	28.6
	5 point	1	2.9

Our observation to date did not reveal the fact that operative procedure increase the atypical cells above 4 point in the circulating blood. Further study will be necessary to substantiate these preliminary findings and clarify the sampling errors involved in the examination.

(文部省科学研究費, 厚生省研究費による)

264. A STUDY ON THE CANCER CELLS IN THE PERIPHERAL BLOOD OF PATIENTS WHO RECEIVED ANTICANCEROUS DRUGS ON RADIATION THERAPY

YOSHIO WADA and SHUNJI HASEGAWA

(Dept. of Internal Med., Nagoya National Hospital)

There have been many papers which reported recognition of the cancer cells in the peripheral blood. The frequency of the appearance, however, is much varied in different authors. This fact may suggest the difficulty which is involved in the identification of the cancer cells in the peripheral blood.

In this study, samples of the venous blood obtained by Seal's silicone method were used to look for the "giant cells", by which abnormal cells that could not be considered hematopoietic origin were meant. We were able to identify the "giant cells" from the peripheral blood in 16 of the 32 patients (50%) with various kinds of cancer.

In 8 patients with primary and 3 patients with metastatic lung cancers, radiation therapy or anticancerous drug therapy was done, anticipating its possible promoting action to the liberation of the cancer cells into the peripheral blood. The samples were obtained from the cubital artery before and after the above treatment to search for the "giant cells". In none of the cases the "giant cells" were found.

265. EXPERIMENTAL STUDY OF THE TUMOR CELLS IN CIRCULATING BLOOD

HIROSHI SAITO and YUKIO OMORI

(Dept. of Surg., School of Med., Niigata Univ.)

A suspension containing 100,000 cells of Walker 256 carcino-sarcoma was injected into the portal vein of 96 rats of Wistar King inbred strain. Material consists of three groups of rats; group 1, control; group 2, grossly hepatectomized (70%) rats; group 3, rats with the blocked reticuloendothelial system by India-ink. Blood samples were collected from femoral, portal and hepatic veins and from the left ventricle on seven different intervals extending till the 14th day after inoculation. Tumor cells stained with a new fluorochrome, N.T.S., previously administered intravenously, were collected by flotation method using Arabic gum solution adjusted to s.g. 1075 for fluoromicroscopic examination.

Examination of the metastatic foci in liver, lung and kidney on paraffin-section revealed that in group 1 and 2 they appear first in the liver on the fifth day of inoculation, then they become conspicuous in the kidney of group 1 and inside the liver of group 2, while in group 3, microscopic foci are recognized in liver on the 3rd day and these develop shortly into extensive metastasis in all of the three organs. Liver retains most of the cells injected into portal vein, in the circulating blood, however, the cells inversely decrease with the appearance of degenerated cells and they increase again along the development of multiple metastasis in the liver. In group 1, the cells decrease sharply and come close to the zero-line on the 5th to the 7th day. In group 3, the down-hill is moderate and goes up again on the point from zero-line. The curve of the group 2 runs roughly between those of group 1 and group 3. Conclusion is that the liver plays a role as a filter against the inoculated cells. In group 1 and 3, its filter capacity is obviously lower than that of the control. Simultaneity of hepatic metastasis and rapid increase of tumor cells in blood suggests an possibility of the liver being the secondary source of metastasis into various organs.

附 議

佐藤春郎：末梢血中の癌細胞出現の場合にはその撒布のもととして肺における腫瘍増殖巢ないし栓塞等癌細胞の存在を意味する場合が多いと思う。

日比野進：流血中には骨髄巨核球、リンパ胚球、網細系に属する細胞等の癌細胞にまぎらわしい細胞が正常人の血液中でも若干出現するものである。本日の臨床の御報告はこれらの異常細胞と癌細胞との同定は全く立派になされているのに敬意を表します。今後腫瘍の back-grounds factors (例えば腫瘍の位置、腫瘍の条件等) により流血中の癌細胞の出現の状況を整理しますとこれまでの混乱しておるとき観を与えておるこの流血中の癌細胞の問題が整理されてくるものと思う。次に異常細胞(癌細胞にあらざる)が癌患者に増加しておることはすでにわれわれは報告しておるが、まことにこのことだけでも興味あることと考えられる。

田崎勇三：流血中の癌細胞の研究に際しては、いつも atypical cell が問題となる。われわれの研究および文献によればこの atypical cell は正常人にはきわめてまれに流血中に出現し、炎症または良性腫瘍においては少数出現するのみである。しかるに癌の場合にはかなり高率のこともあり出現しないこともあってまちまちであった。

すなわち組織診断との関係をみると Adenoca. papillotubulare では atypical cell 0 で癌細胞 12.5% に対して、tubulare medullare では前者 6 で後者 25.0%, acinosum では前者 2 で後者 25.0%, gelatinosum では前者 0 で後者 33.3%, scirrhosum では前者 5 で後者は 33.3% の高率を示した。

すなわち組織学的所見の差異によって、流血中の atypical cell の出現率がまちまちであり、さらに癌細胞の出現率に差異が現われることは興味ある所見である。

266. EFFECTS OF NITROMIN ON CANCER CELLS CIRCULATING IN THE BLOOD (STUDIES WITH ASCITES TUMORS OF RAT)

HIROSHI SATOU, SHIGERU MIZOTA and HIDETARO TOKUYAMA

(Dept. of Surgery, Med. Inst. of Sasaki Foundation)

It was investigated whether Nitromin, an anti-cancer drug, may have inhibitory effect on the intravascular emigration of rat ascites tumor cells (Yoshida sarcoma cells and ascites hepatoma, AH 13 cells) which had been inoculated into the right thigh muscle of rats.

After Nitromin was injected respectively into the abdominal cavity of the tumor bearing rats as many as 0.5 mg/day during 2-4 days (1-2 mg in total), the blood was aspirated from the local vein, V. cava inferior, at the 5th day after the inoculation in Yoshida sarcoma group, at the 10th day in AH 13 group. The blood was centrifuged at 2,000 rpm for several minutes, employing "Hematocrit tube", and white colored superficial layer of the sediment was smeared and stained with Giemsa solution. By the same procedure without the Nitromin injection the control trial was carried out.

The results were as follows: The frequency of appearance of Yoshida sarcoma cells in the local vein decreased markedly by the Nitromin injection while in the corresponding tumor cells were seen these degenerative change as the swelling of the cytoplasm and nuclei, the multinucleation and karyorrhexis. However these morphological changes were never observed in the control group. In the AH 13 group, the emigration of tumor cells were inhibited so markedly that the tumor cells could be scarcely detected in the local vein. The results suggested that the anti-cancer agents could prevent the metastatic formation not only by inhibiting the emigration of tumor cells but also by the injurious effect on the emigrating tumor cells so far examined.

XIII. Immunity

267. STUDIES ON THE CROSS IMMUNITY AMONG THE HOMOLOGOUS TUMOR CELLS

ISAMU USUBUCHI

(Dept. of Path., School of Med., Hirosaki Univ.)

The rats cured of one of such ascites rat tumors as Yoshida sarcoma, Hirosaki sarcoma, Usubuchi sarcoma, hepatoma 130 and hepatoma 7974 showed an intense resistance against the intraperitoneal retransplantation of the same tumor as well as of the different tumor. Tumor cells retransplanted disappeared from the ascites within 24 hours in most cases. Sometimes they continued to grow during several days. Death after prolonged course of tumor growth was observed in rare occasions, especially in cases of retransplantation of hepatomas.

The cross immunity among the homologous tumor cells seems to show that at least one part of the antigenicity of the homologous tumor cells is common. As this common part of the antigenicity must be included in every generated tumor cells, it is expected that the human tumor cells may be affected by the antibodies produced by its own cells, especially in cases of cancer chemotherapy.

(文部省科学研究費による)

268. STUDIES ON THE CROSS IMMUNITY BETWEEN HETEROLOGOUS TUMOR CELLS

ISAMU USUBUCHI, MICHİYOSHI SUGAWARA and JUNNOSUKE YOSHIDA

(Dept. of Path., School of Med., Hirosaki Univ.)

1) SN 36, a mouse lymphosarcoma, was transplanted intraperitoneally in hybrid rats in succession. The tumor cells disappeared from the ascites about 7 days after the transplantation in general, but nodular tumor mass could be palpable intraperitoneally subsequently during 5~7 days in most cases. After that, most rats healed from the transplanted tumor, but some of them especially young animals weighing under 70g, died from the heterologous tumor growth.

2) The rats thus cured of the heterologous tumor were then transplanted with

either Hirosaki sarcoma or Usubuchi sarcoma, an ascites sarcoma of the rat. Most of the rats died of the retransplanted homologous tumor, but some of them were cured of the tumors.

The data seem to show that the mouse and rat may be near relatives and therefore, the antibody against mouse tumor can inhibit the proliferation of rat tumor to a certain degree. (文部省科学研究費による)

269. THE EFFECT OF ACTIVE IMMUNIZATION ON DEVELOPMENT OF MAMMARY TUMORS IN C₃H (JAX) NICE (ADDITIONAL REPORT)

SHINZO ISOJIMA

(Dept. of Gyne., Med. School, Osaka Univ.; Roswell Park Memorial Inst.)

This paper is a preliminary report of an attempt at active immunization in C₃H mice with spontaneous breast cancer. A part of this work was presented at Annual Meeting of American Association for Cancer Research (1958) by author, Ruth M. Graham and John B. Graham, and the additional results are given in this paper. Two separate experiments are reported here. First one on autoimmunity in subtotal and total excision of tumor and second on prevention of tumor by active immunization technique. Spontaneous C₃H tumor mice (Jax) with tumors of 1 cm or more in diameter were used. The tumor was excised, leaving behind a small portion about 2-3 mm. in diameter. The tumors in another group were completely removed. The tumor was homogenized with saline, or ground in a mortar with saline. It was then emulsified with Freund adjuvant and injected into the mouse from which the tumor had been removed. Tumor growth was observed weekly. C₃H mice injected with a homogenate of their own tumor plus Freund adjuvant showed no suppressive effect on tumor growth, and no remarkable preventive effect on tumor recurrence. Autoimmunization appeared to prolong the survival time and the tumor size at death was larger than in the control group. Young C₃H virgin female mice (Jax: 6-10 weeks old) were injected with fresh pooled C₃H tumor plus Freund adjuvant. Each mouse was injected with a total of 0.1 ml. of this material and received a second and third injection. As a control group, young mice were injected in the same manner and with the same volume of Freund adjuvant containing no tumor homogenate. Immunization with pooled fresh C₃H tumor tissue plus Freund adjuvant appeared to decrease the tumor incidence at the 10th and the 12th month but the suppressive effect

became unclear at the 15th and the 18th month. Injection of Freund adjuvant alone appeared to decrease the tumor but not as much as the homogenate plus Freund adjuvant.

附 議

白淵：私は 1956 年札幌での癌学会で、ラットの腫瘍組織で反復高度免疫した場合にこの動物はその後 Methylncholanthrene 発癌の時期を若干おくらせることを報告しました。

ただいまのお話で発癌を抑制した因子は、乳癌組織であるか、またはそれに含まれると考えられる virus であるか。

相沢：自然発癌にも免疫学的予防が起り得るとすれば、その効果の特異性が興味ある問題となると思います。

河村謙二：腫瘍よりの材料および Adjuvant などによる演者の注射の場合、腫瘍動物に何等かの反応性症状またはアナフィラキシー様の症状はなかったか。

追加、何等症状の出ないというような場合は矢張りそういうものの注射による腫瘍発育抑制あるいは注射物と量によってはその促進等の作用は少いのは当然と思う、良好な免疫関係が成立していないためである。私はこのようないろいろの点を腫瘍移植の場合の他正常組織の同種または異種移植の実験結果から考えてきた。

石橋幸雄：Ehrlich 癌細胞を静注して肺腫瘍を作製して、マウス同種肺組織 Adjuvant の影響を検索したが、2 週前に前処置した群は腫瘍形成が著明に阻止された。非特異的な腫瘍抑制因子があることを示すものと思う。

磯島：「免疫原になるのは、milk factor かまたは腫瘍特異物質と考えるか」の質問に対し、現回の所 milk factor の濃度も不十分ですしまた Virus free のマウスを使用しなかったので immune tolerance の問題も絡んでくるため、効果があるとしても、どちらのためかは分らない。

磯島：「腫瘍発生が少い動物群について、アナフィラキシー状態のような傾向は認められなかったか」に対する答

血中の circulating antibody をみるため、tanned hemagghetenation test を行いましたが認めるべき抗体価はありませんでした。が Finks は、lyophilized tumor + Freund Adjuvant を用いた実験で Anaphylactic になるよう証明しています。

磯島：「自分の実験でも Freund Adjuvant 注射で腫瘍の非特異的抑制効果を認めた」に対する答

私も対照として、Freund Adjuvant 注射のみでなく、無処置群をとっておくべきでした。最初無処置群を持っていましたが、途中事故のため全部死亡し、ここで引用することができないのは残念です。

270. IMMUNOLOGICAL ANALYSIS OF THE PARAPROTEIN FRACTIONS IN THE SERA FROM PATIENTS WITH MALIGNANT TUMORS

NOBUTATSU TAKAYANAGI and YASUO MIYAKE

(Clinical Lab. of Kurashiki Central Hospital)

In previous communications, the presence of the abnormal antigenic components in the sera from cancer patients was established using the immunoelectrophoretic method. The experiments to be reported in this paper provide additional informations concerning the nature of the paraprotein fractions in the sera of patients bearing malignant tumors. Details of the experimental conditions of the immuno-

electrophoresis will be discussed in another paper.

In the reaction patterns of the cancerous sera against homologous absorbed antisera, several new precipitation lines were found in β - and α -globulin portions. The thin slices of agar layers completed precipitation patterns were stained with some specific reagents, i.e. Amido black, Sudan black B, Oil red O and PAS reagents. These results indicated that the glycoprotein and lipoprotein were involved in the constituents of the cancer antigens in the serum. These antigens were demonstrated more strongly in advanced cancers than in early cases and had no relation to the histological classification and the original organs of cancers. When the sera from patients with various diseases were analysed with absorbed anticancer sera, some samples of inflammatory or metabolic diseases gave a few faint lines in α -globulin portion. Therefore, it may be assumed that some of the specific cancer antigens cross react with the proteins increased in other metabolic disorders.

The immunoelectrophoretic patterns of the serum from malignant lymphoma against the specific absorbed antiserum gave a sharp precipitin arc in the site of β_2 -globulin, which could not be detected in normal and most of cancerous sera. In the leukemic sera, as described previously, specific antigens were found in β_2 -globulin fraction. It is possible to recognize the differences between the immunoelectrophoretic patterns of leukemic sera and those of cancerous materials.

It is very suggestive to find out the specific paraproteins in each serum of malignant tumors by application of the precipitin tests in gelified media.

附 議

相沢：抗原として腫瘍組織を用いた場合の結果がありましたら、お知らせ下さい。

高柳：人体癌組織における免疫学的分析については現在まとめてはいないが、マウスエーリツヒ癌、ラットの DAB 肝癌について肝組織に異常抗原が現われることをたしかめ一昨年の本学会で報告している。

高柳：抗癌血清の作製は癌患者血清、癌性腹水およびその分画 (III, IV: Cohn) それぞれ材料として家兎を免疫する方法は Adjuvant 法を用いて蛋白総量 200~300 mg 量を 2 回筋注する。多くの抗血清より特異性のつよい抗体価の高いものを正常人血清で吸収して使用した。

271. STUDIES OF THE SERUM PROPERDIN LEVELS IN TUMOR-BEARING PATIENTS AND ANIMALS (II)

MORIMASA SEKIGUCHI, KIYOSUKE OKADA, MICHIO MATSUKURA
and YUKIO ISHIBASHI

(Dept. of Surgery, Inst. for Infect. Dis., Univ. of Tokyo)

The amounts of properdin were determined in 116 sera by the bacteriophage-neutralization assay. PhN₅₀ units of 46 sera from cancer patients ranged from less

than 1 to 42 with a mean value of 13.9, whereas in 12 normal sera, from 3 to 28 with a mean value of 11.9. In the majority of the cancer sera, the properdin levels were the same as those of normal sera with the exception of one serum showing a high level of 42. In 28 sera from patients with pulmonary tuberculosis, the mean value was 12.5 and in 30 sera from patients with miscellaneous diseases other than cancer and tuberculosis, it was 12.2. In these four groups, the properdin levels fell in the same range and the mean values were almost the same, though the value for cancer sera was slightly higher than others. Therefore, as far as the properdin assayed by the phage technique is concerned, it does not favor the view that the cancer patients have low properdin titers.

Seventy to 190 mg of Mitomycin-C in all was given in a few days to advanced cases for the treatment of cancer revealed a rapid fall in properdin levels together with severe leucopenia. It might be thought that the decrease in properdin could afford a favorable conditioning for the bone marrow transfusion thereafter, however, it could bring about the enhancement in bacterial infection of the patients. The local and intravenous administration of Mitomycin-C, 10 to 30 mg as a single dose, however, did not show the remarkable fall in properdin levels.

In some cases of cancer, the properdin titers were determined in series by the phage-neutralizing technique as well as a modified zymosan assay. No parallelism was found between the two.

The experiments with rabbits showed that the immunization with phage increased in the phage units of properdin, though the zymosan units were still unchanged. The fact seems to indicate that the specific phage-neutralizing antibody influences the results of the phage assay.

In general, bacterial and viral infections are prone to occur in the late stages of cancer patients, and thus produces specific as well as cross-reacting substances to the phage. This could be an explanation for the increase in properdin levels in those cases.

From the data presented, it might be considered that the serum properdin levels does not reflect the prognosis of the patients. Furthermore, it seems reasonable to conclude that resistance of the host against cancer is mediated through some factors other than properdin system.

附 議

赤井貞彦：1) d-d 系マウスに Ehrlich 癌を移植する前後に zymosan 1 回 (1 mg) を腹腔内に注入した。移植 4 日前に zymosan を投与した群は移植後 11 日を経ても腹水なく血清 properdin 値も増加を示した。移植 3 日前から移植 1 日後までの間に投与した群はいずれも腹水を生じ properdin 値もわずかに増加または減少を示した。

2) 胃癌患者の血清 properdin 値は健康人の平均値 (14.8 単位) より低く、平均 12 単位であった。また癌病巣の拡がりの大なるものほど低下の傾向を示した。

3) 胃手術後に血清 properdin 値は1時低下する。術後3日目をもっとも著明で7日目にはほぼ回復する。また手術侵襲の大なるものほど低下はいちじるしい。

4) 胃癌患者で保存血輸血後血清 properdin 値の上昇がみられた。

相沢：① 従来の zymosan assay で一定の差があるといわれているもの〔癌患者と健康人、結核症の場合の増殖型と浸出型等〕について特に zymosan assay と phage assay を平行検索してお示し下されば有難い。

② 赤井氏の附議に対し追加

zymosan の腫瘍移植におよぼす影響を示す実験の一部と示説 273 で発表した。

272. STUDIES ON THE RELATIONSHIP BETWEEN SERUM PROPERDIN AND CANCER (III) EFFECT OF VARIOUS DRUGS ON THE DECREASE OF THE SERUM PROPERDIN LEVELS CAUSED BY ANTITUMOR AGENTS

KUNZO ORITA and KAZUTADA MIYAKE

(1st. Dept. of Surgery, Med. School, Okayama Univ.)

Previously the author reported that when an anti-carcinogenic agent is administered to normal and cancer bearing animals *in vivo*, there occurs a transient fall in serum properdin level. This time investigations have been carried out to determine whether or not the combined administration of various drugs with carcinogenic agents (mainly Thio-TEPA) can prevent the fall in serum properdin level. The drugs concurrently used are essential amino acid mixture, vitamins (B_6 , pantothenic acid, folic acid, orotic acid, K), adenine, and cystine. Using mainly rabbits, 1 mg./kg./day Thio-TEPA has been administered with the drugs mentioned above consecutively for seven days, and changes in serum properdin level have been pursued. The results show that the combination of 2 ml. amino acid, 10 mg./kg. pantothenic acid, 10 mg./kg. orotic acid, and 5 mg./kg. vitamin K with Thio-TEPA raised properdin level and that the level is elevated even in the cases given each of these drugs alone.

Next, when pantothenic acid is injected after the intraperitoneal administration of Ehrlich ascitic cancer cells into mice, the span of life is clearly prolonged in the group receiving the dosage over 5 mg./mouse.

Taking a note in the action of pantothenic acid similar to that of ACTH, 5 mg./day of ACTH or cortisone is injected into rabbits for 7 days, and the properdin level falls immediately after the first injection and it recovers to pre-injection level 2-3 days after withdrawal of drug. When 0.5 g./day GABA having the action like ACTH is administered in the same manner, properdin level rises after the first

injection, and even after the drug withdrawal it remains higher than the pre-injection level. When ACTH, cortisone or GABA is administered to Ehrlich ascitic cancer mice, a slight prolongation of the life span has been found in the cases given 10 mg./mouse/day GABA. Moreover, there can be found hardly any inhibitory action on respiration *in vitro* with pantothenic acid, ACTH, cortisone, and GABA. From these findings it is assumed that anti-carcinogenic action of pantothenic acid and GABA is due to re-inforcement of natural humoral defence mechanisms. In addition, there can hardly be recognized any parallel relationship between changes in leucocyte count (the one of factors for natural cellular resistance) and that in properdin level.

273. THE EFFECTS OF ZYMOSAN ON HUMAN TUMOR GROWTH IN THE CONDITIONED RAT

MIKI AIZAWA

(Dept. of Path., School of Med., Hokkaido Univ.; Sloan-Kettering Inst. for Cancer Research)

It has been shown that zymosan affects host-tumor relationship to a certain degree. The present report deals with the effects of the agent on growth of a human tumor, HS#1, in the Wistar rat conditioned with X-ray and cortisone.

Single doses of either 15 mg/kg or 350 mg/kg of zymosan were injected intraperitoneally at different times from 5 days prior to 1 day after the irradiation followed by the subcutaneous tumor inoculation 1 day later. The tumor growth autopsied on the 12th day was compared with that in the control animal.

Tumor growth in rats injected with a low dose of zymosan 5 days before X-ray was inhibited. Enhanced growth was investigated in animals treated with the agent of a low or high dose 8 hours prior to the irradiation. Neither significant inhibition nor enhancement was found by zymosan administration regardless of the doses given 1 day before, just before, or 1 day after X-ray.

These results were discussed from the view-point of the conceivable relationship between serum properdin and zymosan in natural resistance. However, since no significant differences of serum properdin levels in animals which bore the enhanced or inhibited tumor growth were observed at the time of tumor implantation, there seemed to be no direct influence of serum properdin on initial growth of the inoculated tumor.

**274. MECHANISM OF CYTOLYSIS OF EHRlich ASCITES TUMOUR
CELLS BROUGHT INTO CONTACT WITH THE NORMAL
HUMAN SERUM (VII) THE NATURE OF THE
HEAT-LABILE FACTOR**

AKIO TOKUNAGA, JUN OKAMURA, SACHIIHIKO KIJIMA,
and GORO KOSAKI

(2nd Surgical Clinic and Institute for Cancer Research, Med. School, Osaka Univ.)

The following data have been reported previously on the mechanism of oncolysis of Ehrlich ascites tumor cells (EATC) brought into contact with normal human serum.

1) After adsorption of the heat stable factor (γ -globulin) by the surface of EATC (1st stage), oncolysis occurs as a result of the joint action of the heat-labile factor and Mg^{++} (2nd stage). 2) In many respects the heat labile factor resembles the complement, but also differs from it in certain ways. For example, guinea-pig serum, which is a source of the complement in the hemolytic system, is not active in the oncolytic system, because of the inhibitors in it.

The following facts were known about the inhibitors in guinea-pig serum.

- 1) It was not inactivated by heat treatment at 56°C for 30 minutes.
- 2) It was not removed by absorption with a large amount of EATC.
- 3) It was non-dialysable.
- 4) It disturbed the interaction between EATC and the heat stable factor.

Moreover :

1) By dialysis against distilled water for 48 hrs. the oncolytic activity was abolished; however the complement activity for the hemolytic system was still demonstrable after this treatment.

2) On mixing the mid piece of the complement (C_1') with R_1 (i.e. a reagent containing C_2' , C_3' , C_4') the hemolytic activity reappeared, but not the oncolytic activity.

3) However, if to this mixture of R_1 and C_1' was added a fraction precipitated by 1.39 to 2.0 M saturated ammonium sulfate the oncolytic activity reappeared.

From the above results it is likely that for oncolysis of EATC a special component is necessary besides the well known 4 components of the complement. This special component was :

- 1) non-dialysable
- 2) heat-labile (destroyed by heat treatment at 50°C for 30 minutes)
- 3) destroyed by dialysis against distilled water (but not against physiological saline).

275. MECHANISM OF CYTOLYSIS OF EHRlich ASCITES TUMOUR CELLS BROUGHT INTO CONTACT WITH THE NORMAL HUMAN SERUM (VIII) THE EFFECT OF NATRIUM IONS

JUN OKAMURA, HIROMU HIGASHI and GORO KOSAKI

(2nd Surgical Clinic and Inst. for Cancer Research, Med. School, Osaka Univ.)

We have already reported in connection with the mechanism of cytolysis of Ehrlich ascites tumor cells (EATC) in contact with the normal human serum, that the inorganic cation, magnesium plays an essential role in cytolysis.

In this report, results of experiments on the effect of sodium on cytolysis are described.

Normal human serum was dialyzed overnight against 0.25 M sucrose solution containing 0.1 M $MgCl_2$, to reduce its Na content to less than 2 mEq/ml. This serum was added to EATC suspended in the 0.25 M sucrose solution. After one hour's incubation at 37.5°C, the cells were examined microscopically.

Very little morphological difference from the control was seen, and notably there was no blister formation or cell-swelling.

However these apparently normal cells could be stained with Azur-II. They were not transplantable into susceptible mice and showed a marked reduction of endogenous respiration. Therefore it was concluded that these treated cells were severely damaged.

When NaCl at a final concentration of 0.95%, was added to this sucrose medium there was a marked change in the cells, so that it is conceivable that, at its physiological concentration, the Na ion is closely related to the morphological changes occurring during cytolysis.

NaCl of 0.95%, at a final concentration was added to cells which had been incubated with dialyzed serum for an hour at 37.5°C. There was little cell swelling or blister formation. Therefore it was concluded that severely damaged cells could no longer actively transport sodium or passively transport water.

276. STUDIES ON THE LOCALIZATION OF THE SPECIFIC ANTIGEN IN TUMOR CELLS

KATSUO TAKEDA, KOKICHI KIKUCHI, KATSUAKI ITAKURA,
and NOBUYUKI TANIGAKI

(Dept. of Path., School of Med., Hokkaido Univ.)

Intracellular distribution of the specific antigen(s) of rat ascites tumors, mainly the Takeda sarcoma and ascites hepatoma gDT-2, was studied.

Investigations were made on transplantation immunity of mice immunized against TCA-treated subcellular fractions of the tumors on normal liver of the rat to intraperitoneal challenge with the tumors.

The inhibiting activity specific for the tumor was most concentrated in the nuclear fraction but was absent in the microsomal and the soluble fraction of the tumor. The mitochondrial fraction was found less active than the nuclear fraction. All the fractions of normal rat liver cells were inactive.

DNA-protein extracted from the tumor cells displayed no inhibiting effect, while the residual substance insoluble in 1.5 M and 0.14 M saline solution inhibited only the same tumor. Both DNA-protein and the insoluble residue of the normal rat liver were inactive.

Similar results as to distinction of tumor antigen from normal cell antigen were obtained also in the case of the DBA induced hepatoma and normal portion of liver from the identical rat.

This fact suggests that the specific antigenicity found does not relate to genetic difference.

The 1.5 M saline-insoluble fraction of whole tumor cells which contained little amount of DNA inhibited the same tumor specifically, whereas the 1.5 M saline-soluble fraction did not in spite of abundant DNA in it. When physiological saline solution was used for the fractionation, the active fraction was also insoluble.

Further investigation of fractionated nuclear component of the tumor cells indicated that the 1.5 M saline-insoluble residue had the specific antigenicity but the 1.5 M saline-soluble supernatant did not.

All these findings indicate that the tumor specific antigen lies most concentrated in the nucleus, especially in its insoluble component but not in DNA-protein.

277. IMMUNOLOGICAL CORRELATION BETWEEN DAB HEPATOMA AND MC SARCOMA INDUCED IN AN IDENTICAL RAT

KATSUO TAKEDA, YUKO TSUJI, KOSHI MARUYAMA
and TSUNEO MAKI

(Dept. of Path., School of Med., Hokkaido Univ.)

As reported in the previous papers, it was suggested that DBA induced hepatoma or MC induced sarcoma of the rat had the independent tumor specific antigen distinct from the species specific antigen common to rats.

In the present studies, the following experiment was carried out in order to solve the problem as to whether, or not, both the hepatoma and the sarcoma specific antigens really existed after complete exclusion of genetic difference of rats in which the tumors were induced.

For this purpose, simultaneous carcinogenesis of two different type of tumor in the same animal was attempted by feeding with DAB and subcutaneous injection of MC. Two pairs of hepatoma and sarcoma which could be referred to as double cancer have been successfully induced in 2 rats among the animals used. To date, 2 strains of ascites hepatoma and 2 strains of subcutaneous solid sarcoma have been successively transplanted.

By use of these tumors as antigens, the immunological correlation between two different types of tumor in one rat was studied.

The present paper reports the inhibiting effect of mice immunized with those tumors upon transplantation of some existing ascites hepatoma and sarcoma of rats.

Mice, heavily immunized with DAB hepatoma or MC sarcoma which antigen was treated with TCA and with Freund's adjuvant was added, showed a tendency to inhibit the takes of the induced hepatoma, existing DAB hepatoma and MC sarcoma similarly.

On the contrary, mice immunized mildly with either of the tumors treated with TCA without Freund's adjuvant, indicated almost no inhibition against the challenge of these two types as normal mice.

Considering various conditions of immunization, such as content of antigenic cells and the challenged tumor cells, it is stated that at present the mice immunized moderately with hepatoma inhibited the challenged hepatoma more conspicuously than MC sarcoma and *vice versa* mice immunized moderately with MC sarcoma inhibited the challenged MC sarcoma more intensively than hepatoma.

From those tendency it is suggested that two different tumors induced in one rat have independent antigenicity from one to the other, although the antigenic difference

between both tumors is not sufficiently apparent yet. The further experiments are now in progress.

278. STUDIES ON THE TUMOR SPECIFIC ANTIGEN USING RADIOIODINATED ANTISERUM GLOBULIN

KATSUO TAKEDA, YUKOU FUKUSHI, NOBUYUKI TANIGAKI,
and KATSUAKI ITAKURA

(Dept. of Path., School of Med., Hokkaido Univ.)

Serum globulin of rabbits immunized with an ascites hepatoma (gDT-2) derived from an inbred rat of Gifu strain was salted out and radioiodinated.

Aliquots of the globulin solution of both pre-and post-absorption either with the normal rat liver (GL) or gDT-2 were incubated with the tumor cells or the live cells *in vitro* and the radioactivity of the globulin combined with gDT-2 cells or GL cells was measured. Although the radioactivity of the globulin prior to the absorption counted higher in a GL-bound globulin than that in a gDT-2-bound globulin, after satisfactory absorption with GL the latter had a little higher counts than the former which radioactivity was as low as the control normal radioiodinated globulin.

Following the complete absorption of the unabsorbed globulin with gDT-2, the radioactivity of both gDT-2 and GL was reduced to the control level.

From this fact, antihepatoma serum seems to contain two component; one is to be absorbed with the liver, the other is to be absorbed only with hepatoma: namely species and organ specific antibody to attach to both hepatoma and liver, and hepatoma specific antibody to attach to the hepatoma alone.

This suggests that hepatoma cell contains less amount of species and organ specific antigen than liver cell and hepatoma specific antigen missing in the normal liver.

279. INFLUENCES OF ADDITIONAL TRANSPLANTATION OR EXCISION ON GROWTH OF THE TAKEDA SARCOMA

KATSUO TAKEDA and TOSHITAKA KANEMOTO

(Dept. of Path., School of Med., Hokkaido Univ.)

A study was made on changes of growth of the Takeda sarcoma inoculated into the susceptible rats by several manipulations; excision of the implanted tumor, ad-

ditional transplantation into either subcutaneous or hematogenous routes.

Subcutaneous tumor growth initiated by inoculation of the cells 10^7 or less reached at the almost same size as a total, regardless number of the inoculum. Neither recurrence nor metastasis was found following early removal within 3 days after inoculation of subcutaneous implant.

When the excision was performed 7 days or more after the inoculation, recurrence and metastasis apparently increased in some cases. Accelerated growth of metastatic tumor caused by removal of the primary tumor was not resulted from dissemination by surgical procedure, but from rapid growth of already established latent metastasis, since additional hematogenous implantation at the time of excision of the subcutaneous mass did not enhance visceral spread.

However, when proliferative focus remained unremoved elsewhere, the increase in metastasis was not seen, and generally the remained focus grew large. When one nodule of double inocula was removed, the nodule left grew as large as did single inoculum, but growth of metastasis in the visceral organs was inhibited. Growth of additional subcutaneous implant within 6 days after the initial subcutaneous transplantation was inhibited, on the contrary the growth of the initial implant was accelerated.

Simultaneous inoculation of the same number of the cells into both subcutaneous and hematogenous routes revealed smaller subcutaneous mass and compensator enhanced spread in the organs as compared with control growth and when the subcutaneous nodule was excised, growth in the organs was more enhanced.

XIV. Case Report

280. THREE CASES OF DOUBLE CANCERS

AKIO KOBAYASHI, SEIICHI OHASHI and MASASHI KANEKO

(Path., Dept. of the Tokyo 1st National Hospital)

Malignant tumors are increasing and also double cancers are noticed from the standpoint of clinicopathology. Recently we have experienced 3 cases of double cancers. These cases are as follows.

Case 1. 72-year-old Japanese male, whose chief complaint was obstinate vomiting. Then giant diverticle of the esophagus was detected by X-ray test in April 1953. Though he had continued medical therapy, he became cachexia, and died in January 1960. After autopsy, one child first sized esophageal diverticle, within which a dove egg sized papillomatous tumor developed was found and showed squamous cell cancer histologically. Simultaneously, a child fist sized tumor, Borrmann IV type, was recognized in the pylorus of the stomach, which showed histologically adenocarcinoma.

Case 2. 78-years-old Japanese female. Since she had taken notice of the left mammary tumor in January 1960, it had grown up to wal-nut size in August. On the other hand she felt decrease of appetite and had vomiting from the beginning of July. Gastrectomy with gastroenterostomy and left mastectomy were performed in August. By operation a small wal-nut sized tumor was observed in the stomach along the small curvature, which was medullary cancer histologically. In the breast we identified a small wal-nut sized solid tumor which showed histologically acinar cancer.

Case 3. 59-years-old female, who had complained of a small tip sized tumor in the skin of the left lateral ankle in April 1956. It showed amelanotic malignant melanoma in histological findings. Soon after new nodules appeared at the subcutaneous portion of the left lower leg one after the other, which were histologically metastases of the lymph nodes. As tumorous defect was pointed out at the gastric street by rentogenological examination in July 1960, the stomach resection was performed in the same month. After the operation a goose egg sized tumor, Borrmann I type, was identified at the same place like case 2, which showed histologically adenocarcinoma. About this case we are following up until now.

All three cases are relatively old year patients and have stomach cancer respectively. These ones are having many instructive points from the pathogenesis of cancer.

**281. AN AUTOPSY CASE OF GASTRIC CARCINOMA SHOWING
EXTREMELY SEVERE GENERALIZED ANASARCA DUE
TO GENERALIZED EMBOLIC ACROANGITIC
CARCINOMATOSIS IN THE DERMIS**

NOBORU TANAKA, WEI-CHA CHEN and NOBUO KURIBAYASHI*

(Dept. of Path., Japan Red Cross Central Hospital; Dept. of Path., Tokyo
Med. & Dental Univ.*)

The present unusual report is that of gastric carcinoma showing generalized embolic acroangitic carcinomatosis, particularly significant in the dermis, which caused an extremely severe generalized anasarca.

Clinical course: The patient was a 43-year-old male who was rather obese and had been hypertensive for the past several years. The edema which had begun from the left leg, gradually extended to the thigh and neck, finally developing into severe generalized anasarca. Body weight reached nearly 100 kg due to increased body fluid. During that period, he recognized a lymphadenopathy in the left axilla, biopsy of which revealed poorly differentiated carcinoma, possible of the adenomatoid type. Primary gastric carcinoma was highly suspected by histological pattern of the biopsy material, as well as by the fluorographical findings. Southey drainage was continued from both thighs and amounted to approximately 30,000 cc. Etiology of anasarca was not clarified, although marked hypoproteinemia was seen which was a sequel of loss of body fluid rather than the cause of edema. Hypothyroidism was also encountered as one of the cause of edema, because of markedly low BMR (-25.2%). The patient expired of glottis edema 9 months after onset of edema.

Autopsy findings: The entire body was extremely edematous and the skin appeared somewhat different from usual edema, characterized by uniformly firm and rather elastic consistency. Cyanotic discoloration was seen, particularly on the upper portion of the body and thigh with scattered patchy hemorrhages corresponding to the sites of the hair follicles. There was no evidence of any external masses nor nodules palpated throughout the skin. A moderately advanced gastric carcinoma was disclosed which showed Borrmann type 3 and mostly consisted of poorly differentiated adenocarcinoma tubulare including focal areas of signet ring cell type. Lymph node metastasis was significant, found even in the superficial nodes. Definite hematogenic metastasis was histologically evidenced only in the sternal bone marrow. Lymphangitic spread of carcinoma metastasis was conspicuous in various organs and tissues, such as the thyroid, kidney, liver and adrenal. The thyroid was extensively infiltrated by carcinoma, metastasized through the lymphatic pathway, which might have reflected in low BMR. Prominent spider-web-like appearance of ectatic lymph vessels

seen all over the bowel serosa, as well as on the surface of the portion of the liver, even on the inner surface of the cysts, which were incidentally found as polycystic disease of the liver and kidney was demonstrated as fairly pronounced embolic lymphangitic carcinomatosis. The lesion possibly displayed an accumulation of chylous ascitic fluid (600 cc). Most striking lesion was that the skin presented considerable dilation of the thin-walled vessels throughout the dermis, from the upper corium to the deeper structure, all of which were occluded by carcinoma emboli and occasionally contained a scanty amount of blood. Most of them were coiled around the small or moderate sized arteries or venous branches. The wall was very delicate with occasional very scarce fibers. Some of them could not be distinguishable from capillaries of blood or those of lymph. No extravascular invasion of carcinoma was observed. Organizing thrombosis was frequent, containing scarce degenerative and inactive neoplastic cells.

Comment: The actual cause of anasarca was apparently due to embolic acroangitic carcinomatosis. Only a few similar cases have been reported in classic German literature by Schierge (Virch. Arch. 237; 129, 1922), Schucker (Virch. Arch. 267; 339, 1928) and others. All of them were described as "lymphangitic carcinomatosis". The exact nature of these vessels of the present case was not apparent, however, several points of histological characteristics of the vessels, above mentioned, suggested the likelihood of terminal blood canaliculi, particularly arteriovenous junction. Although reconstruction through serial sections gave still more weight to this impression, to demonstrate the exact communication of all this vasculature was particularly difficult and complete explanation is still lacking. Only when fully established complete serial sections are attainable, would it be possible to distinguish between the discrepancy of blood and lymphatic vessels. Thus, the present case is believed to be an illustrative case to investigate a pattern of terminal vessel communication of the skin.

282. AN AUTOPSY CASE OF CARCINOSARCOMA WHICH ORIGINATED IN THE STOMACH

MASAHISA KYOGOKU, TAMOTSU OKUKUBO and SHIGEHISA AOKI
(Path. Inst., Faculty of Med., Kyoto Univ.)

The so-called carcinosarcoma is recognized by us as a kind of malignant-mixed tumor in which the epithelial and non-epithelial components both have equal malignancy, with the exception of collisionstumor. In general, it happens in the middle or old age and chiefly originated in the uterus, oesophagus and lung. It develops

slowly and there's scarcely any metastasis. According to Saphire & Vass among a number of reported cases (153) just three or four were the real carcinosarcoma, and most were nothing else but the highmetaplastic or anaplastic carcinoma and collisions-carcinoma. The case reported here is undoubtedly the real one and is supposed to have originated in the stomach, and which will be the tenth case in this country and we have never seen such complicated case as ours so far as we know.

A 49-year-old Japanese male, about three years ago, was subjected to a subtotal gastrectomy under a diagnosis of stomach cancer, which was histopathologically a well-differentiated carcinoma cylindroepitheliare. But a part of its stroma showed a rather fibromatous pattern and a round cell infiltration. After three years of a symptomless period he had a relapse. At the beginning, he complained of a full feeling in his stomach and followed by nausea, vomiting, jaundice, ascites and finally he went into a coma before dying. The autopsy revealed four neoplastic tumors near the stomach. The larger two were on both stump of the stomach, that is on the wall of cardia and duodenal ampulla. The smaller two were seen on the colon transversum. The one on the duodenal wall compressed the ductus cholelocus and portal vein, and consequently the cholangiolitic liver cirrhosis and ascites occurred. Their histological pattern is complicated; and almost same in the four tumors; it is composed of adenocarcinoma cuboepitheliare, rhabdomyosarcoma with marked striation, leiomyosarcoma and fibrosarcoma. These completely mingle each other but there is a distinctive boundary between the epithelial and non-epithelial components.

Inferring from these facts, we considered as follows: Three years ago, in the stroma of adenocarcinoma there would have been germinal tissue, which would have a potency to differentiate into a number of mesenchymal tissues, and by that time metastasis would have been located in the stump of stomach. And then, in these three years, that stromal germ would develop into rhabdomyosarcoma, leiomyosarcoma, fibrosarcoma and mingle with the original adenocarcinoma. If the carcinomatous component had the higher malignancy, it would completely restrain the growth of sarcoma, however, in this case, the well-differentiated carcinoma would allow the formation of those complicated malignant-mixed neoplasm. Those which were on the colon would have metastasized from the one on the duodenal ampulla. But the possibility of a collision tumor which originated from the same organ would be left over.

283. TWO AUTOPSY CASES OF LEIOMYOSARCOMA DERIVED FROM THE GASTROINTESTINAL TRACT

HISANDO KOBAYASHI and SATOSHI KANO

(2nd Dept. of Path., School of Med., Nagoya Univ.)

Case 1. A 70-year-old male who complained of nausea and upper abdominal discomfort for 7 months before his death. In admission, a firm peanut-sized nodule was found at inner side of the right elbow. Histological examination of this nodule revealed the spindle cell sarcoma of undetermined origin. Chief autopsy findings were as follows:

1. A comparatively firm child-head-sized tumor of grayish-white in color, spreading to the radix mesenterii, with ulcer formation (2×2 cm in size) at pars descendens of the duodenum. No metastatic foci in other organs.

2. A dove's-egg-sized abscess encapsulated with mesenteric wall and serous membrane at the ileocecal region. Histological findings were as follows:

The tumor tissues were composed of large elongated cells which had abundant eosinophilic cytoplasm and large elliptic shaped nuclei with uneven hyperchromatin, occasionally with basophilic nucleoli. Cytoplasm were stained yellow with Van Gieson method and purple with Mallory method. In many areas, a feature of highly irregular palisading arrangement of these cells was seen and in some areas whirl-like, neurinomalike, and round small cell sarcoma like patterns were found.

In metastatic nodule somewhat different picture was noticed, a mixed pattern of small spindle cells and polygonal giant cells of large irregular shaped nuclei, accompanied with round cell infiltration was noticed.

Case 2. A 54-year-old male who complained of vomiting and epigastric pains for 2 months before his death. Chief autopsy findings were as follows:

1. Double palm-sized relatively firm gastric tumor of irregular dish like shape at the cardinal region spreading to the greater curvature, with a ulcer covered with necrotic mass.

2. Several numbers of round metastatic foci of little finger tip in size in the liver.

3. Adhesion between the posterior wall of the stomach and the spleen.

4. Small abscesses in both kidneys.

Histological findings were as follows:

The tumor tissue showed typical leiomyosarcoma patterns in many areas but benign myoma like pattern and hydropic degenerative features in some areas.

Metastatic foci in the liver showed neurinoma-like pattern.

284. A BIOPSY CASE OF VASCULAR TUMOR AT THE POSTERIOR RECTAL WALL RESEMBLING TO BONE GIANT CELL TUMOR

MASASHI KANEKO, SEIICHI OHASHI and NORIKO FUKUSHIMA

(Path. Dept., Tokyo 1st National Hospital)

There are many opinions about origin of the bone giant cell tumor. We had experienced one biopsy case of vascular tumor resembling to bone giant cell tumor.

A 22-year-old Japanese male who had complained of lumbago, slight abdominal pain and difficult defecation during about two months before operation, was diagnosed as tumor at the posterior rectal wall 3cm oral from the anus. But he had no remarkable changes at os sacrum by X-ray examination and blood figure were almost normal. Other physical findings were also normal. In our hospital almost all tumor tissue could be extirpated with the rectum from the sacrum with which this tumor loosely adhered. The patient was quite well after operation and the tumor was double fist sized, brownish black colored and spongy. Histologically, there were many capillaries and small vessels filled with many erythrocytes. At many places of the hemangioendothelial layers, we could see transitional changes from endothelial cells to giant cells of epulis type. Stromal cells had spindle shaped or oval nuclei but did not show any malignant findings such as atypia and mitosis. There were also many scattered same giant cells between these stromal cells. In the stroma there were many slit-like or lacuna containing these giant cells and a few erythrocytes, which showed ring formation of argentophile fibers by silver impregnation. We thought that these giant cells were made up from hemangio-endothelial cells in the course of vascularformation. There were also only a few osteoid formation in the stroma. These tumor cells could form rare osseous tissues like those in giant cell tumor of bone. Accordingly, we thought that this tumor was, in another words, giant cell tumor arisen from angioblastic mesenchymal cells in the neighborhood of the sacrum, even if it showed quite resemblance to hemangioblastoma.

As our conclusions, this case is indicating an important proof that some kind of bone giant cell tumors are angiogenic origin.

285. AN AUTOPSY CASE OF MALIGNANT MESENCHYMOMA OF THE OMENTUM ASSOCIATED WITH DERMATOMYOSITIS

NORIO MATSUOKA, MASAYOSHI SAGARA*, YOSHIHIRO HAMADA*,
and MASAYOSHI HAMADA*

(Dept. of Path., School of Med., Yokohama Univ.; Numazu Municipal Hospital*)

A male, aged 23, in good health until the onset of the present disease (Feb. 1960) visited the hospital with the chief complaint of the abdominal distension. Physical examination at that time revealed marked ascites and many tumor formations in the abdominal cavity. On the 21st day of hospitalisation Mitomycin was administered in the abdominal cavity and on that day multiform exudative erythema appeared on the skin of the elbow and knee joints of both sides which disappeared after 5 days. At the 37th day of hospitalisation, however, diffuse erythrodermic eruption appeared which through dexamethasone administration disappeared after 2 weeks leaving pigmentation on the skin.

The patient expired after 102 days since the onset of the disease.

Autopsy findings: Gross, multiple tumors, the size of pea to chestnut, yellowish gray in color were found on the large omentum. The tumor dissemination was limited to the surface of peritoneal covering and visceral involvement absent. Accordingly primary tumor was considered that it developed from the serous membrane of the abdominal cavity.

Microscopic findings: Generally the tumor was markedly vascular and composed mostly of the mesenchymal cells. Individual cells were spindle shaped and had clear cytoplasm and hyperchromatic round nuclei. Somewhat alveolar arrangement was noted but it did not belong to carcinoma such as metastasized from the other organs like stomach and pancreas. Silver staining revealed fine network which enveloped each tumor cell. Conclusively malignant mesenchymoma was diagnosed developing from the peritoneal tissue. Skin lesion which appeared in form of multiform exudative erythema or diffuse erythrodermia was confirmed as dermatomyositis related with the above-mentioned mesenchymoma. Dermal disease associated with malignant tumor of the visceral organ are reported on the occasion.

Author's case belonged to mesenchymoma and to carcinoma which showed that this tumor was a little rare case.

286. A CASE OF NON-CHROMAFFIN PARAGANGLIOMA IN THE RETROPERITONEUM

RYOZO SANO and NINORI HANADA*

(Aomori Prefect. Central Hospital; Surg. Clinic, Aomori Communication Hospital*).

A forty-two-year old woman was admitted to the Aomori Communication Hospital in April 1960 with a mass in the upper abdomen.

At the time of admission, physical examination revealed a firm, roughly surfaced, thumb-sized nodule in the right upper quadrant of the abdomen which gave rise slight pain upon pressure.

Temperature, pulse and blood pressure were normal. Routine laboratory studies showed no abnormality.

At operation, the tumor formed a circumscribed ovoid mass 3.0 cm in diameter in the retroperitoneum, located between Aorta and V. cava inf., over the 2nd lumbar vertebra. The nodule was easily removed while it adhered to these vessels. Liver, stomach, intestines and pancreas were normal. Postoperative course was good. Grossly, the tumor was egg-sized, well encapsulated and the cut surface was solid, elastic soft, homogeneously reddish brown.

Microscopically, the tumor cells arranged in cluster, alveoli, or perithelially. The majority of neoplastic cells were spindle shaped or polyhedral, with finely acidophilic granular cytoplasm and pale staining round or oval nucleus. Mitotic figure was not seen, but occasionally multinucleated cells were noticed. In parts, indistinct cytoplasmic boundaries appeared to form a syncytium. Many neoplastic cells were directly contacted with the thin walled capillaries.

In some areas, adenoma-like pattern was prominent. It was remarkable in this case, that abundant nerve bundles entered into the stroma. Structurally these bundles were not different from normal one. Ganglionic cells were seen in it. Reticulin fibrils did not extend intercellularly. Neither fat nor glycogen was exhibited in the cytoplasm. The chromaffin reaction was negative. By Bodian's silver stain, argyrophilic granules were not demonstrated.

As a conclusion, this tumor is a typical paraganglioma histologically, although it may be uncertain whether this tumor arises from Zuckerkandle organ or not.

288. A CASE OF SURGICALLY REMOVED OSTEOGENIC SARCOMA OF THE URINARY BLADDER

YOSHIO KONDO

(1st. Dept. Path., Med. School, Nagoya City Univ.)

(Dept. of Dermato-Urology, Nissei Hospital)

A 77-year-old man was admitted to Nissei Hospital complaining of pollakisuria and hematuria associated with radiating pain to anal portion for one and a half years.

After admission, urinary bladder tumor and prostate hypertrophy were confirmed.

On opening the bladder, three variable sized tumors (5, 3 and 2 cm. in diameter) were located in the bladder neck wall.

The tumors were pedunculated, superficially ulcerated, greyish-yellow in color and elastic soft in consistency.

The tumors were resected, the recurrence of them were thoroughly cauterized.

Histologically the tumors tissues contained osseous as well as cartilagenous tissues. The tumor cells were irregular in shape and size (polymorphism.) Multinucleated giant cells and mitosis were numerous. These findings may be regarded as an osteogenic sarcoma.

289. AN AUTOPSY CASE OF PRIMARY MAST CELL LEUKEMIA

KUNIIHIKO SUGA and MAKOTO HAMANO

(Dept. of Path., Kyoto Prefect. Univ. of Med.)

A 14-year-old girl with episode of metrorrhagia and increasing weakness was hospitalized on Sep. 16, 1957. A hemorrhagic tumor was found at the anterior vaginal wall and the malignant tumor was suggested. Blood count revealed, red cells 1,820,000; hemoglobin 43%; white cells 29,000 with 45% metamyelocytes. The diagnosis of acute leukemia was made. The patient was treated with transfusions and Nitromin (50 mg/day, with two days interval, total doses 1,000 mg). Bone marrow tap on Sep. 30 disclosed the increase of basophile granulocytes up to 30%. She expired on Dec. 2, 1957.

At autopsy liver, lymph nodes, kidneys, spleen and bone marrows revealed involvement of the leukemic infiltration, in addition to the grayish-green tumor of anterior vagina, which was non-fluorescent. Histology showed extensive leukemic infiltration, extramedullary development of myelogenous granules of the tumor cells in both infiltration and vaginal tumor demonstrated positive peroxidase reaction, metachromasia by toluidine blue staining, PAS positive, non-digestive by hyaluronidase. These findings confirmed a diagnosis of primary mast cell leukemia.

290. AN AUTOPSY CASE OF ERYTHROLEUKEMIA

KIYOSHI TERAOKA (Dept. of Path., School of Med., Chiba Univ.)

A 51-year-old man was admitted to our hospital on March 7, 1960 with the chief complaints of anemia, weakness, and fever.

There was no enlargement of liver, spleen, and lymph nodes. A total white cell count was 10,400 and a red cell count 1,150,000. The hemogram and myelogram on March 8., were as follows:

Hemogram; Myeloblasts 21.6%, promyelocytes 8.8%, myelocytes 5.6%, metamyelocytes 1.2%, juvenile forms 6.4%, neutrophilic polymorphonuclear leukocytes 14.4%, basophilic leukocytes 0.8%, lymphocytes 7.6%, proerythroblasts 0.8%, megakaryoblasts 9.6%, macroblasts 6.0%, normoblasts 12.4%.

Myelogram: Proerythroblasts 17.0%, macroblasts 13.3%, normoblasts 22.6%, megakaryoblasts 29.4%, mitotic cells 2.1%, myeloblasts 5.3%, promyelocytes 3.3%, myelocytes 1.5%, metamyelocytes 1.2%, juvenile forms 1.2%, neutrophilic polymorphonuclear cells 1.4%, lymphoid reticulum cells 3.8%.

Clinically erythroleukemia was strongly suggested. The patient died on May 5, 1960 under the condition of shock due to the intestinal hemorrhage.

An autopsy disclosed the bone marrow of sternum, vertebral bodies and femur having muddy, red brown appearance. Histologically, it was difficult to identify sinusoids, because of the intense infiltrations of atypical blastic cells of both erythrocytic and leukocytic cell series in the stroma. Spleen weighing 90 g, had a thickened capsule and firm, muddy, and grey-red cut surface. Histologically, it showed the red pulp which was diffusely infiltrated by the leukemic and atypical erythroblastic cells and accompanied by small anemic infarcts in contrast with the atrophic lymph follicles. Liver was slightly enlarged, weighing 1,375 g, and demonstrated diffuse intralobular leukemic cell infiltrations as well as some patchy aggregates. In the ileum wall were multiple up to thumb-tip sized ulcers with hemorrhage, the base of which was diffusely infiltrated by the leukemic cell. In addition to these organ involvements, leukemic infiltrates were found in the tonsils, lungs, kidneys, and testicles.

From the above mentioned clinical and autopsy findings, it may be interpreted and summarized that in this case neoplastic processes took place not only in the leukocyte cell series but also in the erythrocytic series.

附 議

小塚貞雄: 約7年前に右乳癌のため、乳房切除し放射線治療を受けていたが、約3年前、偶然胃痛を発見され胃全摘手術を受けた。その後約2年後、赤白血病に罹患した一例を剖検した。

剖検上、比較的細胞質に富んだ赤白血病細胞が全身諸臓器に彌漫性に浸潤していた。

291. AN AUTOPSY CASE OF EMBRYONAL RHABDOMYOSARCOMA IN THE LEFT AURICULAR REGION

KAZUO HIZAWA and NAOMICHI INUI
(Dept. of Path., School of Med., Tokushima Univ.)

A 6-year-old boy. A rapidly growing tumor of the left auricular portion had been noticed in July 1959. After radiation therapy (^{60}Co , 7400 r), surfical biopsy performed in Nov. 1959 revealed a well differentiated rhabdomyosarcoma. Total excision was not feasible and the patient died in Jan. 1960.

At autopsy the tumor, 440 g and $12 \times 17 \times 7$ cm in size, was found to be situated chiefly in the left auricular region infiltrating the zygomatic, mastoid, temporal and retropharyngeal portions. The zygomatic bone was almost completely, the temporal and sphenoid bone was partly destroyed by the neoplasm. Cut surface was yellowish white and soft edematous. Foci of small necrosis and bleeding were also observed. Metastases were found in the right lung and pleura. Microscopically, both bioptic and autoptic materials were composed of small oval or spindle cells mingled with many differentiated rhabdomyoblasts. The small undifferentiated cells presented small dark stained nucleus and scarce cytoplasm. There were numerous round or oval, thin band-like and multinucleated giant cells rich in eosinophilic cytoplasm which showed fine fibrillar texture. The nuclei of giant cells were bizarre in form and showed at times tandem wise arrangement. Cross striation and glycogen granules were clearly demonstrated in these cytoplasm.

Portions of the autoptic material were composed largely of the undifferentiated small round or oval cells with scanty cytoplasm and gave the impression, to a certain extent, of the round cell sarcoma. In these areas the cells were generally compact, but at times they were very loosely dispersed. In general, this neoplasm was almost devoid of collagenous fibres except in the perivascular area, while fine delicate reticulin fibrils were abundant throughout the tumor tissue. Fibrils came direct in contact with each cells, frequently giving an appearance of radial projection from each cells. At times the fibrils put the cells in row. In the well differentiated portions the fibrils ran adherent and parallel with the long band-like cells.

292. AN AUTOPSY CASE OF BRANCHIOGENIC CARCINOMA

TATSUYA OKAMOTO and NOBUTOSHI KOBAYASHI

(Dept. of Path., School of Med., Chiba Univ.)

An autopsy case of branchiogenic carcinoma originated in the right cervical portion of a 65-year-old man is presented. He noticed swelling of the right cervical portion about 4 months prior to the admission, which gradually increased in size during the last two months. It grew up to the size over man's fist without causing any subjective symptoms except slight pain. And then overlying skin was ulcerated due to the tumor invasion. While he was receiving radiation therapy, the necrosis developed extensively and died suddenly from excessive bleeding.

Pathological findings: The child's head sized tumor having the overlying skin which was ulcerated in two portions was seen and the central part of the tumor showed necrosis and the common carotid artery was exposed in the basis of the ulcer. The tumor extended superiorly to the right lobe of thyroid, r.submaxillary gland and parotid and inferiorly to the r.supraclavicular area. For histological examination, they showed picture of squamous cell carcinoma. There were carcinoma cell nests, which were densely packed together with typical pearl formation, and the carcinoma cell themselves were polyhedral in shape and a few number of mitotic figures, most of which were asymmetrical and bipolar. In the section taken from the tumor tissue of the level of the hyoid bone was found a slender cystic cavity lined by squamous epithelium, presumably a segment of congenital cyst encircled by the tumor tissue. In addition to the above mentioned findings, were found a tissue malformation in the capsule of liver, which was consisted of only bile duct and connective tissue, as well as multiple small cysts in the kidneys.

Comment: This case is considered to be a branchiogenic carcinoma because of the following reasons. There were no primary tumor in any other organ, no evidence of invasion from the thyroid, submaxillary and parotid glands, while a cyst was found in a section at the level of the hyoid bone of the tumor. In addition, this case had congenital malformations in the liver and kidneys.

291. AN AUTOPSY CASE OF EMBRYONAL RHABDOMYOSARCOMA IN THE LEFT AURICULAR REGION

KAZUO HIZAWA and NAOMICHI INUI

(Dept. of Path., School of Med., Tokushima Univ.)

A 6-year-old boy. A rapidly growing tumor of the left auricular portion had been noticed in July 1959. After radiation therapy (^{60}Co , 7400 r), surfical biopsy performed in Nov. 1959 revealed a well differentiated rhabdomyosarcoma. Total excision was not feasible and the patient died in Jan. 1960.

At autopsy the tumor, 440 g and $12 \times 17 \times 7$ cm in size, was found to be situated chiefly in the left auricular region infiltrating the zygomatic, mastoid, temporal and retropharyngeal portions. The zygomatic bone was almost completely, the temporal and sphenoid bone was partly destroyed by the neoplasm. Cut surface was yellowish white and soft edematous. Foci of small necrosis and bleeding were also observed. Metastases were found in the right lung and pleura. Microscopically, both bioptic and autoptic materials were composed of small oval or spindle cells mingled with many differentiated rhabdomyoblasts. The small undifferentiated cells presented small dark stained nucleus and scarce cytoplasm. There were numerous round or oval, thin band-like and multinucleated giant cells rich in eosinophilic cytoplasm which showed fine fibrillar texture. The nuclei of giant cells were bizarre in form and showed at times tandem wise arrangement. Cross striation and glycogen granules were clearly demonstrated in these cytoplasm.

Portions of the autoptic material were composed largely of the undifferentiated small round or oval cells with scanty cytoplasm and gave the impression, to a certain extent, of the round cell sarcoma. In these areas the cells were generally compact, but at times they were very loosely dispersed. In general, this neoplasm was almost devoid of collagenous fibres except in the perivascular area, while fine delicate reticulin fibrils were abundant throughout the tumor tissue. Fibrils came direct in contact with each cells, frequently giving an appearance of radial projection from each cells. At times the fibrils put the cells in row. In the well differentiated portions the fibrils ran adherent and parallel with the long band-like cells.

292. AN AUTOPSY CASE OF BRANCHIOGENIC CARCINOMA

TATSUYA OKAMOTO and NOBUTOSHI KOBAYASHI

(Dept. of Path., School of Med., Chiba Univ.)

An autopsy case of branchiogenic carcinoma originated in the right cervical portion of a 65-year-old man is presented. He noticed swelling of the right cervical portion about 4 months prior to the admission, which gradually increased in size during the last two months. It grew up to the size over man's fist without causing any subjective symptoms except slight pain. And then overlining skin was ulcerated due to the tumor invasion. While he was receiving radiation therapy, the necrosis developed extensively and died suddenly from excessive bleeding.

Pathological findings: The child's head sized tumor having the overlining skin which was ulcerated in two portions was seen and the central part of the tumor showed necrosis and the common carotid artery was exposed in the basis of the ulcer. The tumor extended superiorly to the right lobe of thyroid, r.submaxillary gland and parotid and inferiorly to the r.supraclavicular area. For histological examination, they showed picture of squamous cell carcinoma. There were carcinoma cell nests, which were densely packed together with typical pearl formation, and the carcinoma cell themselves were polyhedral in shape and a few number of mitotic figures, most of which were asymmetrical and bipolar. In the section taken from the tumor tissue of the level of the hyoid bone was found a slender cystic cavity lined by squamous epithelium, presumably a segment of congenital cyst encircled by the tumor tissue. In addition to the above mentioned findings, were found a tissue malformation in the capsule of liver, which was consisted of only bile duct and connective tissue, as well as multiple small cysts in the kidneys.

Comment: This case is considered to be a branchiogenic carcinoma because of the following reasons. There were no primary tumor in any other organ, no evidence of invasion from the thyroid, submaxillary and parotid glands, while a cyst was found in a section at the level of the hyoid bone of the tumor. In addition, this case had congenital malformations in the liver and kidneys.

PRESIDENT'S ADDRESS

DR. YUZO TAZAKI (Cancer Institute Hospital)

Ladies and Gentlemen! It is a great honor to say a few words at this occasion of the General Assembly of our Japanese Cancer Association which has grown up to be one of the most prominent scientific organizations in our country.

Cancer is a most formidable enemy of mankind and we are set to annihilate it. Our efforts have been directed to this aim from all possible aspects. The size of our general assemblies has been expanding year after year, and about 300 papers have been read during the present sessions. Both fundamental problems involved in the mechanism of cancer cells or a portion of a single cell, and improvement in the practical treatment of human cancers were discussed. Many progresses have been reported. It is a pleasure for me to say that our Association is contributing continuously and very much to the world's knowledges of cancer.

However, the way is still long and we must collaborate in the common aim of achieving the solution of many problems remaining.

We are going to have in our program three special lectures this afternoon. It has been very fortunate for us, especially to be able to invite Dr. Sarah E. Stewart from the National Cancer Institute of the United States. As perhaps all of you know well, Dr. Stewart has recently succeeded in isolating the polio virus and introduced an entirely new epoch in the field of experimental cancer. She has been kind to come over and will give you a summary of her intensive studies on this particular subject.

The other subject of the special talks will be the genesis of carcinoma of the stomach. Two speakers were chosen among many Japanese workers to present some recent data of their respective studies on the development of gastric cancer which is one of the most important subjects in this country so far as cancer of the Japanese people is concerned. I hope the choice is good and our audience will be much profited by the special lectures.

POLYOMA VIRUS AS A MODEL FOR STUDIES ON THE VIRAL ETIOLOGY OF HUMAN NEOPLASMS

SARAH E. STEWART

(Laboratory of Viral Oncology, National Cancer Institute,* Bethesda, Md.)

Until the present decade, except for discoveries which showed that viruses cause certain malignant tumors in fowl, proof supporting the infectious theory of cancer was slow in developing. It was not until 1935 that a virus-caused cancer was demonstrated in mammals. At this time Rous *et al.* (1) showed that the Shope papilloma virus scarified on the skin of domestic rabbits produced papillomas which were frequently transformed to malignant tumors. They were unable, however, to transfer the malignancy by cell-free filtrates. In 1936 Bittner (2) discovered in strain C₃H mice a naturally transferable mammary tumor virus that is transmitted by milk to the nursing offspring. These few successes stood out among a great many failures in the effort to find viruses in mammalian cancers, and interest in the possibility waned.

Then in 1951 Gross (3) described the induction of leukemia in newborn strain C₃H mice by means of cell-free extracts of tissues from strain AKR mice with spontaneous leukemia. That same year I attempted to confirm Gross' findings using C₃H mice of the Heston subline. One mice failed to develop leukemia; instead, at 8 to 10 months many of them developed tumors in the parotid glands. This tumor had not been hitherto reported. Using the same procedure we were able to accelerate or induce leukemia in (C₃H/Hen × AKR)F₁ hybrid mice. In 1953 (4) we reported on an agent recoverable from the leukemic tissues of mice which we believed produced both parotid gland neoplasms and leukemia; the type of tumor induced being dependent on the host. Later it was shown that there were 2 viruses involved, one which caused the leukemia and the other the parotid gland tumors. Even a third virus which was believed to cause sarcomas was added (5).

Our attempts to demonstrate a tumor inducing agent in the parotid gland neoplasms by injecting tumor cell-free extracts into newborn mice failed. However, when extracts or cell suspensions from the tumors were inoculated into primary tissue cultures of monkey kidney or of minced mouse embryo we were able to show the release of a tumor-inducing virus in the culture fluids (6-8). Fluids from these cultures produced tumors in 85 to 100 percent of mice injected with it. And it

* National Institutes of Health, Public Health Service, U.S. Department of Health, Education, and Welfare.

induced over 20 different kinds of tumors in the mice, some having as many as 8 different kinds of primary tumors. These neoplasms involved most of the glands of the head and neck, all of the salivary glands, the lachrymal glands, the mucous glands of the sinuses and trachea and the thyroid glands (Figures 1-8). The thymus was frequently transformed into an epithelial tumor which arose in the medulla, apparently from Hassels corpuscles (Figures 9-10). Contradicting the rule that tumors of the mammary glands develop only in female mice, because of the excitation by estrogenic hormones, the virus engendered mammary adenocarcinomas in males as well. (Figure 11). These, however, are different histologically from the ones induced by the milk agent (Figure 12). Tumors of the lungs are usually of the type involving the mesothelium. They were observed only in the Swiss mice (Figure 13). Epidermoid carcinomas involving the hair follicles often covered the entire body surface (Figures 14-15).

The virus not only produces carcinomas, but also sarcomas and hemangiomas. Mice often develop multiple osteogenic sarcomas involving most of the skeleton. Sarcomas of the kidneys and subcutaneous tissues also were found (Figures 16-18). The relatively massive doses made possible by tissue culture exposed a virulence in the virus that is camouflaged by its more covert action in nature.

The questions as to whether we were dealing with one or many oncogenic viruses was raised. Since the virus causes a cytopathogenic effect on mouse embryo tissue culture, we were able to plaque the virus and thus demonstrate that we had only one virus, and that it was capable of producing the large spectrum of neoplasms. This is shown in table 1. Because of the many tumors that the virus can induce we proposed the name "polyoma."

Besides producing many tumors in a given strain of mice polyoma virus was found to cross mouse strain barriers and even species barriers in other rodents. Hamsters, rats and rabbits are also susceptible to tumor induction when inoculated with polyoma virus (8-11). In rabbits the tumors are subcutaneous and resemble fibromas. They regress after a short time. In the hamsters and rats the neoplasms are sarcomas and hemangiomas. These occur at many sites in the hamsters and in the kidney only in the rats. Hamsters are more susceptible to the virus than mice. Tumors may develop within a few days. Hamster tumors are shown in figures 19-21.

Among its many interesting biological properties one which distinguishes polyoma virus from other mouse oncogenic viruses (Bittner milk agent and leukemia virus) is its high antigenicity (12). Antibodies to the virus appear in the circulation of animals inoculated with it whether they develop tumors or not. Mice with large induced tumors will have high antibody titers. Even mice not injected with virus but housed in the same quarters as the inoculated animals develop antibodies (13,

14). Latent infections with the virus as shown by the presence of circulating antibodies have been found in a high percent of the mice in certain colonies (15). The virus has been shown to spread by saliva and excreta (15). The antigenic response provoked in the host would account for the rarity of spontaneous tumors in infected colonies. Antibodies are passed to the offspring by nursing mothers through their milk.

Another property of polyoma virus is its ability to produce a hemagglutinin which agglutinates washed red blood cells from various species (16). The inhibition of hemagglutination by antibodies is a quick and reliable way for titrating sera and milk for antibody content. Antibody titers of mouse sera and milk as determined by the inhibition of hemagglutination are shown in tables 2 and 3.

Polyoma virus has been shown to replicate in the nuclei of infected cells causing a ballooning degeneration. By electron microscopy such nuclei have been shown to be filled with non-capsulated virus particles 27 m μ in diameter (Figure 22). We have shown that the active constituent of this virus is deoxyribonucleic acid (DNA) (17). Early after infection with the virus many cells with ballooned nuclei may be found in different organs. These are especially numerous in the kidneys. Later when cellular proliferation becomes prominent fewer cells are present with replicating virus. In grafts of mouse tumors even after many passages virus is still demonstrable as shown by the presence of serum antibodies in the host carrying the graft and by recovery of virus when the tumor is cultured. This, however, is not true of the hamster tumors. It is impossible to recover virus from the hamster tumor grafts or to demonstrate antibodies in the host carrying the graft. It is possible that the virus DNA becomes incorporated in the cell chromosomes and thus is no longer recoverable by any of our known methods of induction (as used in studies of bacterial viruses).

The experiments with polyoma virus have provided a model for studies with human neoplasms. Attempts have been made to recover oncogenic viruses in tissue cultures inoculated with extracts and concentrates made from human tumors. The results have been discouraging. Several investigators (18-21) have reported tumors in mice and other laboratory animals after inoculation with tumor extracts. But it is now generally felt that the induced tumors are probably the result of activation of latent oncogenic viruses in the host.

Similar experiments which we have carried out with human tumor material in tissue culture made from human embryo mince, hamster embryos, mouse embryos and human amnion, from which fluids were injected into newborn hamsters are of interest because of the high incidence of unusual tumors and lesions which we have observed in our inoculated animals. However, essentially the same types of tumors were observed in control hamsters which received supernatant fluids from the uni-

noculated control cultures. The results which we have recently reported are given in tables 4 and 5 (22).

Of the numerous types of malignancies observed a lesion of the uterus is of greatest interest as this has not been reported either as a spontaneous lesion or as one induced in any experimental animal. It is believed to be of trophoblast origin. Early in our work it was found only in the group of experimental animals. Forty-five percent of the females less than 12 months old died from this lesion. Later it was also found in the young females that were inoculated with the control cultures. It was first observed at post mortem examination of a 4-month old pregnant female which had died spontaneously. The peritoneal cavity was filled with blood and had several normal looking fetuses which appeared to be 1-2 days from parturition. The left horn of the uterus had ruptured but the right remained intact and contained masses of varying sizes. The serosal surface was covered with hemorrhagic projections 2-3 mm in size (Figure 23). On opening the right uterine horn it was found to contain macerated fetuses ranging in size from 0.5 cm to 1.5 cm. The hamster also had an enlarged spleen. Of those animals that died during pregnancy, in many death could be attributed to severe hemorrhage through the vaginal canal. All the hamsters with the uterine lesion had macerated fetuses; in some instances only one, in others all were macerated. In those where all were macerated it was not possible to estimate the time of gestation; but most of the females appeared to be near parturition at the time of death. All uteri examined histologically had massive extravasation of blood throughout the myometrium. Hyalinization of arterial walls, organized thrombi and blood vessels with bizarre giant endothelial cells were found in the muscularis. The blood vessels with the giant endovascular cells extended to the serosa and in 2 instances giant cells had replaced the serosal cells covering the uterus. Figures 24-26 show sections of uteri with the characteristic lesions.

DISCUSSION

Since the hamsters which received the control tissue culture fluids developed essentially the same types of tumors as the ones which received the inoculated cultures the results are difficult to evaluate. Hamster colonies have been described as being relatively free from spontaneous tumors (23-25). Certain of the tumors which we observed, however, have been reported. Fortner (26) observed gastrointestinal adenocarcinomas, intestinal polyps, adrenal cysts, liver cysts, hepatomas, cardiac thrombi, renal lesions, reticulum cell sarcomas, malignant melanomas and an occasional tumor of other types in his hamster colony. His animals consisted of an experimental group which had received injections of bile and other substances and a group that remained uninoculated. His finding with the various groups were similar except for possible potentiation of carcinogenesis in some of the animals which

received human bile.

Spontaneous epidermoid carcinomas in hamsters have not been described. In our animals fourteen squamous cell tumors of the esophagus or forestomach and 2 squamous cell tumors of the skin were observed in 247 experimental animals which have been examined. None were seen in 110 of the control animals which received fluids from uninoculated cultures. Since the groups are not comparable in number, it is necessary to study more animals before it can be assumed that the epidermoid carcinomas resulted from the inoculum given.

The uterine lesions which we have observed appears to be of a type not previously described in hamsters. Orsini (27) has studied in detail the normal gestation in this animal and has described and classified trophoblastic giant cells and endovascular cells observed from the immediate post-implantation period through placentation, parturition, and the first three post-partum weeks. To the primary and secondary giant cells that are discharged with the placenta at parturition, which had been described by others, she has added a tertiary giant cell and an endovascular cell both of trophoblast origin, which are found deep within maternal tissue and maternal vessels. Early in pregnancy the tertiary giant cells in the sheathed arteries of the decidua basalis. Later they are found within the arteries of the mesometrium and appear to form the actual lining of some dilated arteries in the upper portion of the mesometrium. They were not observed in the myometrium. The tertiary giant cells remain within the arterial system into the third post-partum week.

In our inoculated animals bizarre giant cells were found in blood vessels throughout the myometrium in which there was also marked extravasation of blood. In 2 instances giant cells extended to the serosal surface of the uterus.

Since we also observed the uterine lesions in the control animals which received supernatant fluids from the uninoculated cultures, a possible explanation for this lesions is the development of tolerance as a result of the tissue culture inoculum. As trophoblast cells are of embryonal origin, they may become more invasive in a host which has been made tolerant because of inoculation at birth with tissue. It is also possible that the lesion is due to an infectious virus that spread to the controls from the experimental animals. The macerated fetuses so frequently associated with the uterine lesion would favor a virus etiology.

Since the incidence of lymphomas, adrenal tumors, liver cysts, and adenocarcinomas appears to be higher in our group of animals than what has been reported for uninoculated hamster colonies, the inoculum given to the newborn hamsters may have also increased their tolerance for their own neoplastic cells. Defense mechanisms in a host may normally destroy potential tumors in many animals.

Table 1. Comparison of types of tumors observed in mice responding to polyoma virus before and after tissue culture plaquing*—percent with specific tumors

Type of Tumor	Plaques virus	Non-plaques virus
A. Salivary glands and other mucous glands	94	96
B. Renal lesions	20	48
C. Mesotheliomas	56	20
D. Epithelial thymomas	25	25
E. Mammary adenocarcinomas in females and males	25	25
F. Subcutaneous sarcomas and hemangioendotheliomas	12	16
G. Bone sarcomas	53	22
H. Renal sarcomas	3	14.5
I. Hair follicle epidermoid carcinomas	3	9
J. Thyroid carcinomas	20	6
K. Adrenal medullary tumors	3	4

* Histological examination of tissues of 331 mice injected with non-plaques virus and 32 mice injected with plaques virus.

Table 2. Inhibition of polyoma virus hemagglutination by mouse serum antibodies* (plus signs denote degree of inhibition)

Mice	Age Mo. when bled	Serum dilutions							
		1/80	1/320	1/1,280	1/2,560	1/5,120	1/20,480	1/40,960	1/81,920
Virus injected in mice 4 weeks old	14	—	—	—	—	—	—	—	±
"	14	—	—	—	—	—	—	±	3+
"	14	—	—	—	—	—	4+	4+	4+
No virus inoculated held with infected mice	21	—	—	—	—	—	4+	4+	4+
"	9	—	—	—	4+	4+	4+	4+	4+
"	19	—	—	—	—	4+	4+	4+	4+
"	19	—	—	4+	4+	4+	4+	4+	4+
"	14	—	—	—	4+	4+	4+	4+	4+
With adrenal tumor graft 114 G**	2	—	—	—	—	—	—	—	1+

* Sera on 12 month old breeders from environment free of polyoma failed to inhibit hemagglutination above 1/80 dilution.

** Transplanted virus induced tumor carried through 114 serial transfers for period of 7 years.

Table 3. Inhibition of polyoma virus hemagglutination by specific antibodies present in strain C3H mouse milk

Source of milk	Age of mice (months)	Milk dilutions							
		1/10	1/20	1/40	1/80	1/160	1/20,480	1/40,960	1/81,920
2 female mice inoc. with virus when newly born	5	—	—	—	—	—	—	±	3+
2 female mice from virus free environment	6	—	—	—	3+	4+	4+	4+	4+
Cow's milk*	—	—	—	—	—	4+	4+	4+	4+

* Calf serum has been shown to contain an inhibitor which prevents tumor induction in mice.

Table 4. Tumors and lesions observed in 12 to 26 month old hamsters which received culture fluids shortly after birth.

Number of animals	160 experimental		67 controls	
	Number	Per cent	Number	Per cent
Lymphomas				
Plasma cell and reticulum cell	29	18	6	10
Lymphocytic leukemia	9	6	3	4
Adrenal tumors	16	10	10	15
Gut lesions				
Acute enteritis	48	30	15	22
Hyperplasia of mucosal glands	33	20	9	13
Polyps	30	20	10	13
Carcinomas	23	14	9	13
Stomach lesions				
Changes at junction of squamous and columnar epithelium	13	8	5	7
Adenomatosis	4	2.5	0	0
Ulcers	12	8	3	5
Liver lesions				
Cysts	33	20	15	22
Hemangiomas	8	5	5	7
Cholangiomas	10	6	0	0
Spleen lesions				
Atrophy	31	20	10	15
Hemangiomas	9	5	2	3
Uterine lesion	6	7 (of females)	1	1.5 (of females)
Uterine tumors				
Fibromas	2	2 (of females)	1	1.5 (of females)
Adenocarcinomas	2	2 (of females)	1	1.5 (of females)
Trophoblast	0	—	0	—
Epidermoid carcinomas				
Esophagus and forestomach	14	9	0	—
Skin	0	—	0	—
Other tumors				
Sarcomas	2	1	0	—
Islet cell adenoma	1	0.6	0	—
Peritoneal mesothelioma	1	0.6	1	1.5
Lung cancer	2	1.0	2	3

Table 5. Tumors and lesions observed in 4 to 12 month old hamsters which received culture fluids shortly after birth

Number of animals	87 experimental		43 controls	
	Number	Per cent	Number	Per cent
Lymphomas				
Plasma cell and reticulum cell	2	2	0	0
Lymphocytic leukemia	1	0.6	0	0
Adrenal tumors	0	0	0	0
Gut lesions				
Acute enteritis	22	25	9	21
Hyperplasia of mucosal glands	11	12	6	14
Polyps	2	2	2	4
Carcinomas	2	2	0	0
Stomach lesions				
Changes at junction of squamous and columnar epithelium	0	0	1	2
Adenomatosis	0	0	0	0
Ulcers	3	4	2	4
Liver lesions				
Cysts	3	3	0	0
Hemangiomas	0	0	0	0
Cholangiomas	1	1	0	0
Spleen lesions				
Atrophy	8	10	4	10
Hemangiomas	0	0	0	0
Uterine lesion	20	45 (of females)	5	20 (of females)
Uterine tumors				
Fibromas	0	—	0	—
Adenocarcinomas	0	—	0	—
Trophoblast	2	4 (of females)	0	—
Epidermoid carcinomas				
Esophagus and forestomach	3	3	0	—
Skin	2	2	0	—
Other tumors				
Carcinoma of head (origin 1/m unknown)	1	1		

REFERENCES

- (1) Rous, P. and Beard, J.W.: Carcinomatous changes in virus-induced papillomas of skin of rabbit. *Proc. Soc. Exper. Biol. Med.* 32: 578, 1935.
- (2) Bittner, J.J.: Some possible effects of nursing on the mammary gland tumor incidence in mice. *Science* 84: 162, 1936.
- (3) Gross, L.: "Spontaneous" leukemia developing in C₃H mice following inoculation, in infancy, with AK-leukemic extracts, or AK embryos. *Proc. Soc. Exper. Biol. N.Y.* 76: 27-32, 1951.
- (4) Stewart, S.E.: Leukemia in mice produced by a filtrable agent present in AKR leukemic

tissues with notes on a sarcoma produced by the same agent. (Abstract). *Anat. Rec.* 117: 532, 1953.

(5) Gross, L.: Studies on the nature and biological properties of a transmissible agent causing leukemia following inoculation into newborn mice. *Ann. N.Y. Acad. Sci.* 68: 245-256, 1957.

(6) Stewart, S.E., Eddy, B.E., Gochenour, A.M., Borgese, N.G. and Grubs, G.E.: The induction of neoplasms with a substance released from mouse tumors by tissue culture. *Virology* 3: 380-400, 1957.

(7) Stewart, S.E., Eddy, B.E. and Borgese, N.G.: Neoplasms in mice inoculated with a tumor agent carried in tissue culture. *J. Nat. Cancer Inst.* 20: 1223-1233, 1958a.

(8) Eddy, B.E., Stewart, S.E., Young, R. and Mider, G.B.: Neoplasms in hamsters induced by mouse tumor agents passed in tissue culture. *J. Nat. Cancer Inst.* 20: 747-761, 1958a.

(9) Eddy, B.E., Stewart, S.E., Stanton, M.F. and Marcotte, M.J.: The induction of tumors in rats by SE polyoma virus embryo tissue culture preparations. *J. Nat. Cancer Inst.* 22: 161-171, 1959b.

(10) Stewart, S.E. and Eddy, B.E.: A review of the biological properties of SE polyoma virus. Presented at the 7th World Congr. int. Soc. Hematology, Rome. 1958.

(11) Eddy, B.E., Stewart, S.E., Kirschstein, R.L. and Young, R. D.: Induction of subcutaneous nodules in rabbits with SE polyoma virus. *Nature, Lond.* 183: 766-767, 1959a.

(12) Stewart, S.E., and Eddy, B.E.: Properties of a tumor-inducing virus recovered from mouse neoplasms. *Perspectives in Virology*, pp. 245-255 (Wiley, London, 1959.)

(13) Stewart, S.E., Eddy, B.E. and Stanton, M.F.: Induction of neoplasms in mice and other mammals by a tumor agent carried in tissue culture. *Proc. Third Canadian Cancer Conference*, Academic Press, N.Y. 1959.

(14) Stewart, S.E., Eddy, B.E., Irwin, M. and Lee, S.: The development of resistance in mice to tumor induction by SE polyoma virus. *Nature*. 1959.

(15) Rowe, W.P., Hartley, J.W., Law, L.W. and Huebner, R.J.: Studies on mouse polyoma virus infection. III. Distribution of antibodies in laboratory mouse colonies. *J. Exer. Med.* 109: 449-462, 1959.

(16) Eddy, B.E., Rowe, W.D., Hartley, J.W., Stewart, S.E. and Huebner, R.J.: Hemagglutination with the SE polyoma virus. *Virology* 6: 290-291, 1958c.

(17) DiMayorca, G.A., Eddy, B.E., Stewart, S.E., Hunter, W.S., Friend, C. and Bendich, A.: Isolation of infectious deoxyribonucleic acid from SE polyoma-infected tissue cultures. I. A preliminary report. *Proc. Nat. Acad. Sci., U.S.*, 1959.

(18) Schwartz, S.O., Schoolman, H.M., Szanto, P.B. and Spurrier, W.: Studies in leukemia. VI. The induction of leukemia in AKR mice by means of cell-free brain filtrates of humans who died of leukemia. *Cancer Res.* 17: 218-222, 1957.

(19) Bergoltz, V.M.: Experimental studies of the etiology of leukemia in men. A review. *Neoplasma Československa Onkologia.* 5: 337-347, 1958.

(20) Burton, L., Friedman, F., Kassel, R., Kaplan, M.L. and Rottino, A.: The purification and action of tumor factor extracted from mouse and human neoplastic tissue. *Trans. N.Y. Acad. Sci.* 21: 700-707, 1959.

(21) Grace, J.T., Mirand, E.A. and Mount, D.T.: Relationship of viruses to malignant disease. Part II. Oncogenic properties of cell-free filtrates of human tumors. *Archives of Internat. Med.* 105: 482-491, 1960.

(22) Stewart, S.E. and Irwin, M.: Possible complications in the use of laboratory animals for studies on the viral etiology of human neoplasms. *Proceedings of the Fourth National Cancer Congress*. In press.

(23) Ashel, R.: Development of tumors in hamsters and rats following the injection of mycobacteria. Estratto dalle "Pubbl. Staz. Zool. Napoli" 28: 12-31, 1956.

(24) Hebermann, R.T. Personal communication.

(25) Smith, W.W.: Personal communication.

(26) Fortner, J.G.: Spontaneous tumors, including gastrointestinal neoplasms and malignant melanomas, in the Syrian hamster. Cancer 10: 1153-1156, 1957.

(27) Orsini, M.W.: The trophoblastic giant cells and endovascular cells associated with pregnancy in the hamster (*Cricetus Auratus*). Am. J. Anat. 94: 273-331, 1954.

All photomicrographs are of sections stained with eosin-hematoxylin.

Figure 1. Six month old strain C₃H/Hen male mouse with bilateral parotid gland tumors.

Figure 2. Strain C₃H/Hen male killed at 4 months of age. It had bilateral salivary gland neoplasms showing the characteristic nodular formation. It also has an epithelial thymic tumor and a renal sarcoma.

Figure 3. Parotid tumor showing the pleomorphic structure characteristic of these neoplasms. The tumors consist of ducts of epithelial origin and of actively proliferating cells of mesenchymal origin, $\times 50$.

Figure 4. Lung with vascular metastasis from a parotid gland tumor, $\times 220$.

Figure 5. Mucous gland tumor of trachea. Histologically similar to the parotid gland neoplasms, $\times 60$.

Figure 6. Tumor arising in left orbit, $\times 35$.

Figure 7. Epidermoid carcinoma of gingiva extending into the mandible, $\times 40$.

Figure 8. Bilateral carcinoma of the thyroid glands, $\times 37$.

Figure 9. Thymus with marked hyperplasia of Hassall's corpuscles and early tumor formation, $\times 200$.

Figure 10. Thymoma involving only one thymic lobe, $\times 40$.

Figure 11. Mammary adenocarcinomas in Swiss male killed at 4 months of age. Mouse also has a renal sarcoma and salivary gland tumor.

Figure 12. Mammary adenocarcinoma in which the architecture is different from the conventional murine mammary tumor, hair follicle lesions overlying the tumors, $\times 40$.

Figure 13. Mesothelioma covering serosal surface of lung, $\times 135$.

Figure 14. Female Swiss mouse with hair follicle neoplasm and parotid gland tumors. Hair removed with a depilatory.

Figure 15. Section through hair follicle tumors showing areas of degeneration and other areas of hyperplasia of the epithelial cells, $\times 35$.

Figure 16. Three month old Swiss male with bilateral renal sarcomas.

Mouse also has a mammary adenocarcinoma and bilateral parotid gland tumors.

Figure 17. Hypernephrotic kidney from a (C3H \times AKR) F₁ hybrid mouse. The parenchymal cells have been completely replaced by the sarcoma, $\times 2$.

Figure 18. Multiple osteogenic sarcomas in Swiss mouse. Tumor in sacrum, innominate bone, skull, mandible, and right foreleg.

Figure 19. Sarcomas in subcutaneous tissues, heart and liver in male hamster killed at 70 days of age. Animal also has liver hemangiomas.

Figure 20. Sarcoma in wall of bowel in hamster, $\times 80$.

Figure 21. Renal sarcoma in hamster with palisading of cells resembling the structure of human neurinoma, $\times 260$.

Figure 22. Electro-photo micrograph of polyoma virus from mouse embryo culture. Intranuclear

virus 27 m μ in diameter. Photograph made by Dr. Leon Dmochowski and Mr. Clifford Gray, M.D. Anderson Hospital, Houston, Texas.

Figure 23. Horn of uterus with hemorrhagic papillary projections extending through the serosa.

Figure 24. Uterine myometrium with massive hemorrhage causing separation of muscle fibers. The hemorrhage extends to the serosa which is edematous, $\times 260$.

Figure 25. Bizarre giant cells in blood vessels of myometrium. Several are multinucleated cells, $\times 320$.

Figure 26. Uterus with giant cells lining the serosal surface, $\times 260$.



Fig. 1



Fig. 2

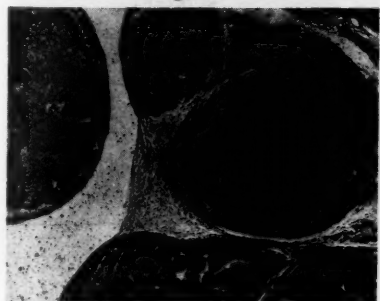


Fig. 3



Fig. 4



Fig. 5



Fig. 6

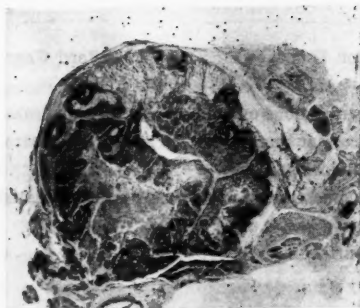


Fig. 7

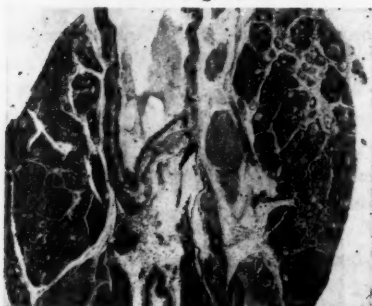


Fig. 8

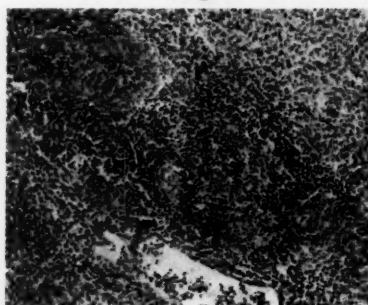


Fig. 9

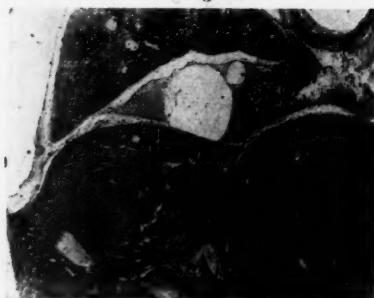


Fig. 10



Fig. 11

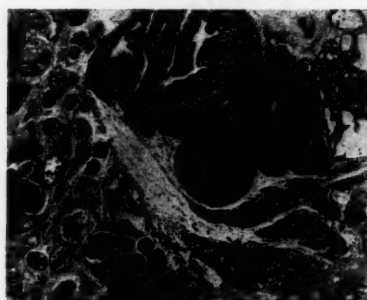


Fig. 12

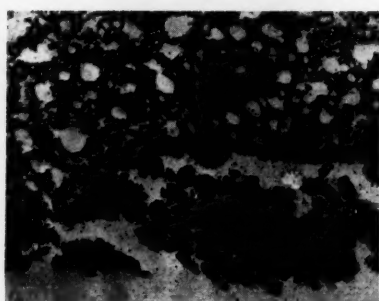


Fig. 13



Fig. 14



Fig. 15



Fig. 16



Fig. 17



Fig. 18



Fig. 19

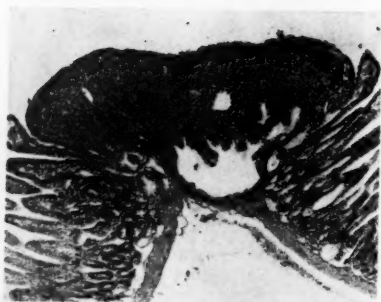


Fig. 20

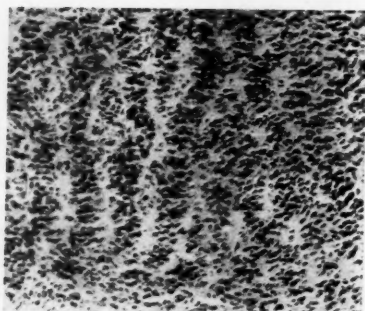


Fig. 21

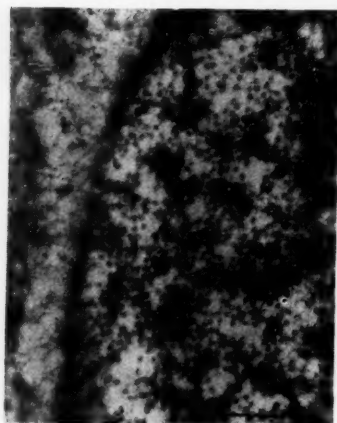


Fig. 22



Fig. 23

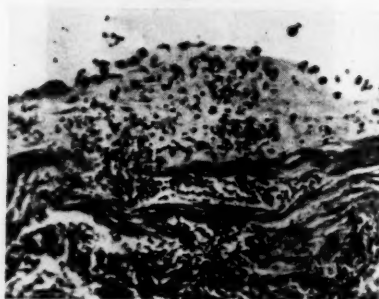


Fig. 24

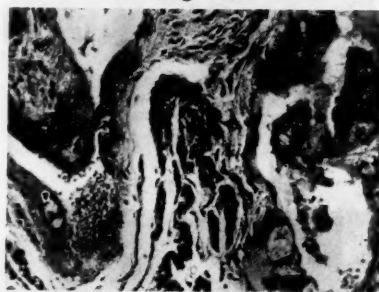


Fig. 25

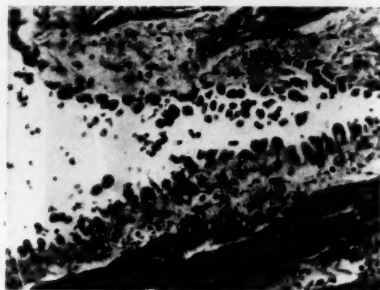


Fig. 26

ON THE POINT OF THE DEVELOPMENT OF STOMACH CANCER

TADASHIGE MURAKAMI

(Department of Surgery, Showa Medical School)

INTRODUCTION

During the past 15 years, I have examined over 2000 stomachs histologically which were removed surgically at the Second Surgical Department, School of Medicine, Tokyo University and the Department of Surgery, Showa Medical School. Half of them were the case of stomach cancer. Out of these specimens of stomach cancer, I have selected about 90 cases of the early stage of cancer and examined their origination histologically.

During the course of histological examination of the serial section of carcinomatous lumina from the large one to the small, I have found the independent carcinomatous lumina by comparing the microscopic photographs of the serial section following one after another. I have named it as a point of development of stomach cancer, since it became nearly a point when it followed to the direction to smaller. Since I have reported them several times at the annual conferences of the Society, presently I will report them as a summary. Many of the figures shown today, were already shown at the annual conferences.

HISTOLOGICAL STUDY

A. ADENOCARCINOMA

Case 1; D.H. 54 yr. male

Date of Operation; April 27, 1950

Clinical Diagnosis; Stomach Cancer

Gross Diagnosis by Operation; Peptic Ulcer

Histological Diagnosis; Stomach Cancer (Ulcerocancer)

A large and elliptical (3.0×2.5 cm) ulcer was found at the stomach angle of the lesser curvature. The early stage of adenocarcinoma was found when the edge of the anterior wall of the stomach, where appeared redder compared to other area, was examined histologically. As the result of the histological examination of all around the edge of the ulcer, cancer was found only a limited part of it. Further histological examination of the serial section of the limited part of it showed that the spreading of the cancer can be divided into two islands, one of which was a large and another was a small. As shown in Fig. 1-A, I have found an isolated carcinomatous lumen in the small island. Its independence from the neighbouring

lumina could be proved by the examination of the serial section. The cells of the carcinomatous lumen were composed by basophilic stained cytoplasm and a large nucleus, in which I have found more than two nucleoli and many mitotic figures. Distinguishable difference between the cells of the carcinomatous lumen and of foveolar (Fig. 1-A') was noticed. Basophilic stained cells in Fig. 1-A showed the same character as ones in lumina in Figs. 1-B and -C. They also showed the same character as the cells in carcinomatous lumina which were definitely proved histologically in separated slides.

Following results were obtained from the above observation :

A) A small carcinomatous lumen (Fig. 1-A) was completely isolated from the another.

B) The basement membrane of the lumen (Fig. 1-A) was clearly shown by silver staining.

C) Only one communication was found between lumina A and A' in Fig. 1 at the point marked with arrow. No other communication of lumen A was found.

D) Lumen A' in Fig. 1 is a regenerated one which has been developed at the edge ulcer. Therefore, carcinomatous lumen A in Fig. 1 is developed at the top of the regenerated lumen. The schema of these findings was shown in Fig. 5-Case 1.

Case 2; I.O. 55 yr. male

Date of Operation; October 3, 1950

Clinical Diagnosis; Stomach Cancer

Gross Diagnosis by Operation; Stomach Cancer

Histological Diagnosis; Stomach Cancer

A linear and large ulcer which was surrounded by erosion, was found at the prepyloric antrum. Many carcinomatous lumina were found at the ulcer. The invasion of carcinomatous lumina was also found in the floor of the ulcer. Fig. 2 shows that lumen A is a glandular part of the lumen A' which has been proved by the serial section. The lumen A' was a pseudopyloric gland which was a regenerated pyloric gland at the edge of the ulcer. The cells in Fig. 2-A were composed of basophilic stained cytoplasm and a large nucleus, in which more than two nucleoli were found. Distinguishable difference between the cells in Fig. 2-A and in the pyloric gland was noticed. The cells in lumina in Figs. 2-B and -C showed the same character as ones in Fig. 2-A. The luminal cells in Fig. 2-C were cancer cells and stood in a row irregularly. The lumen A in Fig. 2 had largest cut surface and was isolated completely from the other, which had been proved by the study of the serial section. Namely, lumen A is cancer which has been developed at the deepest part of the pseudopyloric gland which has been regenerated at the edge of the ulcer. The schema of these findings was shown in Fig. 5-Case 2.

Case 3; H.G. 37 yr. male

Date of Operation; August 20, 1949

Clinical Diagnosis; Stomach Cancer

Gross Diagnosis by Operation; Stomach Cancer and Polyps

Histological Diagnosis; Stomach Cancer and Polypocancer

The Borrmann III type stomach cancer was found at the prepyloric antrum of the removed stomach. Two polyps, top of which had become already carcinomatous lumina, were found at the anterior wall of the stomach and quite independent from the cancer. Mixture of carcinomatous lumina and normal gastric fundic glands was noticed at the peduncle of the polyps (Fig. 3). The cells in lumen A in Fig. 3 were composed of basophilic stained cytoplasm and a large nucleus, in which two nucleoli were found. The lumen A showed only one connection to the lumen A', which was a branch of foveola (A'') of gastric fundic gland (Fig. 3). The cells in lumen A showed the same character as ones in lumina B and C. The lumina B and C showed irregular forms and cells in those showed the same character as ones of the carcinomatous lumina at the top of the polyps. Therefore, the lumen A is the carcinomatous lumen like lumina B and C. It was proved by the serial section that lumen A was completely isolated from the surrounding carcinomatous lumina and distinguishable border was found between carcinomatous cells (A) and normal gastric foveolar ones (A'). Namely, carcinomatous lumen A sits at the top of the branch A' of the foveolar (A'') of the gastric fundic gland.

The location of the lumen A was shown in Fig. 5-Case 3a.

Fig. 4 is another section of a peduncle of polyp. Here also many independent carcinomatous lumina (E and F) were found as shown in Fig. 3. The lumen D was located at the top of the lumen D' which was a branch of foveola (D'') of a gastric fundic gland. Furthermore, lumina D and D' showed the largest cut surface of the branch in this slide. The cells in lumen D were composed of basophilic stained cytoplasm and a large nucleus, in which two nucleoli were found, and showed distinguishable border from the gastric foveolar cells (D'). It was found by the study of serial section that the lumen D was completely independent from the other lumina. By silver staining examination, I have found that the basement membrane was intact. Namely, the lumen D was a small carcinomatous lumen which was located at the top of foveolar branch of the gastric fundic gland.

The schema of these findings was shown in Fig. 5-Case 3b. From the four schemas in Fig. 5, the following findings were proved. The early stage of adenocarcinoma at the edge of ulcer was developed at the top of lumina (Case 1 and 2). In case of polypocancer (Case 3a and 3b), the early stage of adenocarcinoma was located at the top of the branch of the gastric foveola. If the branches can be thought to grow from gastric foveola, developments of both of ulcerocancer and polypocancer were started from the top of growing lumina. Therefore we believe

that the carcinomatous lumina develops from the development center of the gastric gland because it situates at the top of growing lumina.

B. INTESTINAL EPITHELIAL METAPLASIA

Case 4a; T.Y. 43 yr. female

Date of Operation; September 22, 1952

Clinical Diagnosis; Gastric Ulcer

Gross Diagnosis by Operation; Stomach Cancer

Histological Diagnosis; Stomach Cancer

A relatively small ulcer which was surrounded by the early stage of carcinomatous lumina, was found by the angle on the lesser curvature of the removed stomach. Fig. 6 showed the smallest group of cells of intestinal epithelia. They were located by the foveola of the gastric pyloric gland and 7 of them stood in row at most. They possessed cuticle edge and goblet cell, and were clearly bordered by the surrounding foveolar epithelia. As shown in Fig. 7, another 5 groups of the independent intestinal epithelial cells were found at the gastric foveola. Namely, the intestinal epithelial cells are easy to develop at the gastric foveola. For their development they did not always need growing branched lumina.

C. DEVELOPMENT OF SOLID CANCER

Case 5; K.K. 58 yr. male

Date of Operation; May 10, 1949

Clinical Diagnosis; Peptic Ulcer

Gross Diagnosis by Operation; Stomach Cancer

Histological Diagnosis; Stomach Cancer

A small ulcer was found on the lesser curvature near pylorus. Near to the cardiac side, meandering erosion was found such as Konjetzny presented. Following to the section along the lesser curvature about 10 of the independent and solid groups of cancer cells were found. By the examination of the serial section, two groups out of them were proved really independent from the neighbouring groups, and one of which is shown in Fig. 8 as a silver staining preparation. The group of cells which has arrow mark in the figure, possessed clear cytoplasm and their nuclei were pressed to the cellmembrane and showed a form of the signet ring cell. The cells were proved to be epithelial ones, since it was stained positively by mucicarmin staining. This group of atypical cells was so large as to fill in 70 slides which were cut at 5 μ . In the figure, the group of cells showed only two communication to the lumina A and B. No other communication was found. The group of atypical cells might be developed from the epithelial cells of lumina A and B. In the figure 8, the group of atypical cells was connected with the small branch A which

belonged to the lumen A'. By the silver staining examination, it was found that epithelial cells of lumen A was connected directly with the atypical cells at the point where the basemembrane of lumen A was destroyed. The group of atypical cells might be developed from the cells of the destroyed part of the basemembrane of lumen A.

In the lumina A and B, many goblet cells were found (Fig. 8). But since they did not possess cuticle edge and paneth cell, they were not the intestinal epithelial cells. Surrounding them, several small scars of ulcer were found. They might be the regenerated lumina.

Case 4b; T.Y. 43 yr. female

This is the same case as shown in case of intestinal epithelial metaplasia. The lumen A is a branch of the lumen A' in foveola of a gastric gland. At the top of lumen A, basement membrane was destroyed and its form became irregularly. Many irregular epithelial cells were found around the top of lumen A. It was doubtful whether those cells were cancer cells or not. At the right hand of Fig. 9, the early stage of carcinomatous lumina were found. Therefore the lumen A is a small gland which is locating by the outside of the cancer *in situ*. The changes which appeared at the top of the lumen might have some connection with the development of cancer, since this kind of changes at the top of lumen had not been known. Furthermore, these changes were almost same as in the case of the development of the solid cancer. It is not impossible to assume that the changes in lumen A in Fig. 9 is a development figure of solid cancer. Namely, the development of cancer is multi-cellular at a development center.

SUMMARY

It was proved that the development of carcinomatous lumina was found at the top of the growing lumina (Case 1-Case 3). This was true in both cases of ulcerocancer (Case 1 and Case 2) and of polypocancer (Case 3). It was also proved that the development of the intestinal epithelia was found in the foveola of gastric gland. The location of the development of cancer is completely different from the one of the development of the intestinal epithelial metaplasia. However, I have no intention to deny that the cancer does not develop from the gastric foveola and intestinal epithelia does not develop from the top of the growing lumina. If the above mentioned theory is true, the development of cancer is multi-centric.

The solid cancer was also developed from the top of lumina when its basement membrane was destroyed as shown in Fig. 8. As soon as the cancer cells were developed, they were located in the tunica propria mucosae in the case of solid cancer. The solid cancer was also developed multi-centrally and it is suggested that cancer might be developed multi-cellularly in the last figure.

Many reports have appeared on the development of cancer already, however this is the first report on it, using the serial section and studied on the point of its development.

ACKNOWLEDGMENT

I wish to express my sincere appreciation to Professors Tamotsu Fukuda, Seiji Kimoto and Ichiro Hirafuku for their constant encouragement and advice. I wish to thank all the members of the Department of Surgery, Showa Medical School, for their cooperation.

LEGENDS FOR ILLUSTRATION

Fig. 1. A is the smallest carcinomatous lumen which is located at the top of A' in case 1 (ulcerocancer).

Fig. 2. A is the smallest carcinomatous lumen which is located at the top of A' in case 2 (ulcerocancer).

Fig. 3. A is the smallest carcinomatous lumen which is located at the top of A' in case 3b (polypocancer).

Fig. 4. D is the smallest carcinomatous lumen which is located at the top of D' in case 3b (polypocancer).

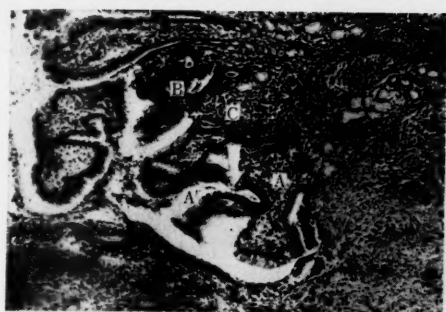
Fig. 5. The schema of the development of carcinomatous lumina (Case 1-3b).

Fig. 6. The smallest group of intestinal epithelial cells in case 4a (arrow marked).

Fig. 7. The schema of the location of the smallest group of the intestinal epithelial cells in case 4a.

Fig. 8. The location of the smallest solid cancer by silver staining method in case 5 (arrow marked).

Fig. 9. The irregular cells at the top of small branch A in case 4b (silver staining).



From above Fig. 1, 2, 3



Fig. 4

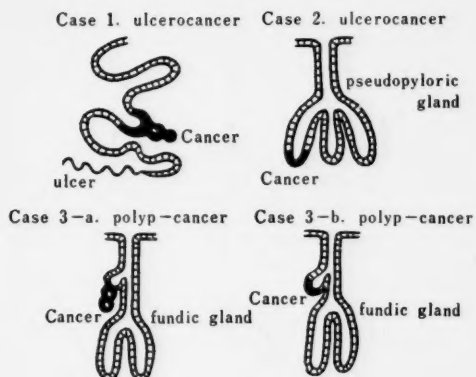


Fig. 5 Diagram of development of adenocarcinoma



Fig. 6



Fig. 8

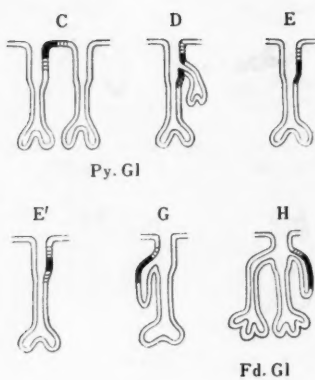


Fig. 7



Fig. 9

ON HISTOGENESIS OF GASTRIC CARCINOMA: A HISTOPATHOLOGICAL STUDY ON THE GENESIS OF SCIRRHOUS CARCINOMA OF THE STOMACH AMONG THE JAPANESE*.

KUNIO OOTA, M.D.

(Department of Pathology, Cancer Institute and
Department of Pathology, Tokyo Medical and Dental College)

INTRODUCTION

One of every three cancers of the stomach among the Japanese belongs to that particular type of carcinoma, called scirrhous carcinoma, whose histogenesis remains still a mystery. Usually this type of cancer does not form a well delineated nodular mass but infiltrates very diffusely until major parts of the gastric wall become involved. Frequent appearance of scirrhous carcinoma among the younger persons, especially female, and its almost absolutely ominous prognosis have attracted much attention.

The histogenesis of gastric cancer in general has been discussed seriously since Konjetzny: some carcinomas are believed to be direct derivatives of adenomatous polypi, some to arise in connection with the presence of chronic ulcers, and many others to develop upon the basis of chronic gastritis. If ever there are such morphological precursors of gastric carcinomas, an attempt at histogenetic analysis of human material, usually obtained by surgical resection or at autopsy, is fertile only in a fraction of available cases, because progression of the malignant lesions has very often obscured the precedent benign changes recorded in loco. But, with a large series of gastrectomy material in hand, a pathologist may be lucky enough to collect experiences in carcinomas in their very early phase of development, and, making use of such accumulated knowledge, he may perhaps explain correctly the histogenesis of more advanced lesions where only traces of precursor changes remain.

In this presentation, the author's attention will be restricted solely to a certain aspect of the problems involved in the histogenesis of the scirrhous carcinoma of the stomach which somehow has escaped the analysis of the previous authors, because, as it appears to the present author, this represents a key point for the whole problem of gastric cancer among the Japanese.

* Aided by the Scientific Research Grant, Ministry of Education.

MATERIAL AND METHOD

In total, 1793 stomachs with cancer resected at the Surgery Clinic of the Cancer Institute Hospital, Tokyo, during a period between 1946 and 1959 comprise the basic material of this study*. All the material were examined routinely in a standard histologic procedure and classified accordingly. Of the stomachs with scirrhous carcinomas, 300 were thoroughly cut down into multiple (30-70) paraffin blocks, parallel to the lesser curvatures, and completely step sectioned. Several staining methods were applied including H & E, Mallory's azan, mucicarmine and silver impregnation.

CLASSIFICATION OF GASTRIC CARCINOMA

Table 1 shows the prevalence of several histological types among the carcinomas examined. The important role played by the scirrhous type is evident: at least about 33% of all material belong to this particular type (Table 1).

Table 1. Histological Classification of Carcinoma of the Stomach

1. Adenocarcinoma papillotubulare	22.2%
2. Adenocarcinoma tubulare medullare	35.3%
3. Adenocarcinoma acinosum medullare	4.7%
4. Adenocarcinoma scirrhosum	32.8%
5. Adenocarcinoma gelatinonodulare	4.6%
6. Adenocarcinoma gelationcellulare	3.3%
7. Adenoacanthoma	0.3%
*Total cases 1743	100.0%

(Cancer Institute, 1960)

CHARACTERISTICS OF BENIGN ULCERATION AS PRECURSOR

The criteria for the diagnosis of an ulcer-carcinoma have been introduced by Hauser. Murakami has tried to expand them and included more cases in the category. But, both Hauser and Murakami have dealt only with such deep ulcerative changes as had once penetrated the entire layers of the proper muscle coat of the stomach (UL-IV) as precursors of cancer. None has yet tried to establish a set of criteria for the recognition of shallower ulcerations which later become superimposed by malignant lesions in loco. In the group called ulcer grade III (UL-III), only the inner layer of the proper muscle coat is reached by the ulcerative process. In the reparative stage, the UL-III represents almost all characteristics of the more authentic chronic ulcer (UL-IV) except the subserosal fibrosis.

The third type, called ulcer grade II (UL-II), represents the chronic or healed states of ulcers which had reached only to the submucous layer. The proper muscle coat remains essentially unchanged, but the mucosal muscle reveals features due to amputation, scarring, and luxurious regeneration, while there is corresponding fibrosis in the submucous layer. The mucous membrane has been completely re-

* By courtesy of the Surgery Clinic, Cancer Institute Hospital (Director: Dr. K. Kajitani).

generated or else shows at least some signs of attempted regeneration. The architectures of the regenerated mucosa are often incomplete as to their glandular differentiation and arrangement. It often shows tufty appearance: the tufts are very frequently slanted towards the center of the previous ulceration and their thickness tapering away in the same direction. Heterotopic glandular regenerates are incidentally observed.

When a cancer originates and grows in loco, many of the above benign lesions, mostly products of reparation, become involved in the cancer and lost almost completely, but some of them may be found remaining at some location if such are extensively searched for by means of step sectioning of the entire material.

CHRONIC ULCERS ASSOCIATED WITH ADVANCED SCIRRHOUS CARCINOMAS

Scirrhou carcinoma over 4 cm in the greatest diameters were deliberately called "advanced" in this study. This group consisted of 424 cases. Of the 424, 284 had grossly noticeable ulcerative changes, irrespective of their malignant and benign nature, while the rest, 140, no apparent defect over their surfaces. The 140 stomachs and additional 65 cases with gross ulceration were cut down thoroughly and histologically examined. Of the total of 205 cases, 112 showed evidences of chronic peptic ulcers which preceded the occurrence of malignant lesions. As listed in Table 2, 43 of them belonged to the rather classical UL-IV and UL-III

Table 2. Preexisting Benign Ulcers and Their Scars As seen in 205 Advanced Cases of Scirrhou Carcinoma

	UL-II	UL-III	UL-IV
Active Ulcers	42	15	23
Scarred Ulcers	27	4	1
Total 112	69	19	24

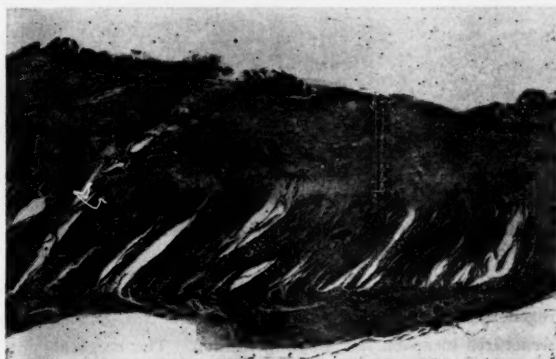


Fig. 1. A classical example of scirrhou carcinoma of the stomach with diffuse infiltration of the entire layers of the gastric wall. Note the remarkably thickened submucosa. Over the surface there is regeneration of the mucosa with its tufty appearances. Scirrhou carcinoma originating in the area of a healed grade II ulcer (UL-II). (Case 0-3879)



Fig. 2 Magnification of the mucosal lesion in Fig. 1. Note the characteristic tufty regenerating mucous membrane and loss of the mucosal muscle. (Case 0-3879)

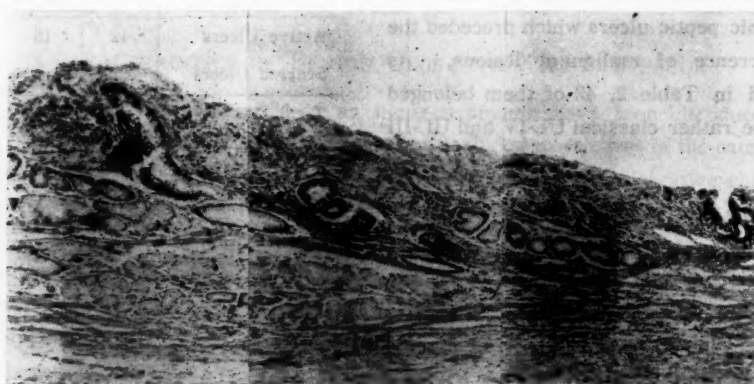


Fig. 3 An early stage of scirrhus carcinoma of the stomach involving the regenerating mucous membrane. Note the arrangement of the glandular structures in peculiar slanting fashion. The mucosal muscle has been partly amputated. The cancer shows isolated cell infiltration of the submucous fibrosis. (Case 0-2531).



ulcers, but the majority, 69, to UL-II, shallower ulcerative changes.

The findings suggest that scirrhus carcinoma of the stomach is frequently associated with previous chronic ulceration, especially shallower one. The results are thought to be very significant, inasmuch as such evidences of precedent benign lesions are brought about in such a high percentage in the examination of the group of advanced cancerous stomachs. The ulcerative changes and scars are observed usually in the center of the cancerous infiltration or slightly eccentric within the involved areas.

CHRONIC ULCERS ASSOCIATED WITH RELATIVELY SMALL SCIRRHUS CARCINOMAS

The scirrhus carcinomas, histologically labelled as such but smaller than 4 cm in their greatest diameters, were seen 68 times in the present series. The age and sex distributions of them are shown in Table 3. There was none in the age brackets below 29 years, and this fact may be explained by the assumption that scirrhus carcinoma tends to spread more rapidly in the younger age than in the older.

When the whole materials were thoroughly examined histologically, 59 cases (86.8%) of them showed evidences of pre-existing benign ulcerative lesions. Here again, although less

Fig. 4 An advanced scirrhus carcinoma in association with an UL-III ulcer which shows regeneration of the mucous membrane in parts. Note the characteristically diffuse infiltration, and also the elevation of the proper muscle coat at the center showing adhesion to the regenerating mucosal muscle. (Case 0-2764)

Table 3. Sex and Age Distribution, Smaller Scirrhus Carcinoma of the Stomach

	-19	20-29	30-39	40-49	50-59	60-69	70-79	Total
Male	0	0	8	12	14	7	1	42
Female	0	0	4	7	7	7	1	26
Total	0	0	12	19	21	14	2	68

impressive, the shallower ulcers are predominant.

In these smaller scirrhus carcinomas, the growth pattern of the cancer is represented in a miniature of the advanced cases: the mucous membrane overlying the ulcer scars or present at the margins of active ulcers is

replaced by infiltrating carcinoma, and the scars, especially prominent in the submucous layers, are infiltrated in such a way that the whole pattern in a cut surface is revealed as a fan-like extension of the cancer through the scars.

Table 4. Preexisting Benign Ulcers and Their Scars As Revealed in Smaller Scirrhus Carcinomas

	UL-II	UL-III	UL-IV
Active Ulcers	14	15	20
Scarred Ulcers	8	2	0
	22	17	20/59

DISCUSSION

One of my collaborators, Takagi, recently reported his examination of 52 cases of mucosal carcinoma of the stomach. None of his cases was associated with the presence of a polyp or advanced carcinoma. Thirty-six (70%) were in conjunction with previous chronic ulceration of benign nature, most of which belonged UL-II. Another colleague, Someya, has studied 60 cases of early ulcer-carcinoma: 27 cases (47%) of his series showed either UL-II or UL-III type of benign ulcerations as the precursor changes. Thus, it is apparent that in many of the early cancers of the stomach their origin can be traced down to the area where shallow peptic ulceration has been present some time.

The early stages of infiltration of the submucous scars were identified in over 50% of the above two series and the histological features were consistent with those of the small scirrhus carcinomas described above. About half of them all showed isolated cell infiltration, occasionally with formation of the signet ring cells. Distinct tubular patterns formed by the infiltrating cells were occasionally observed. As has been pointed out by Nagayo and Takagi, intestinal type of metaplasia of the adjacent gastric mucosa was very slight in the cases with isolated cell infiltration.

Thus, there seems to be evidence that a carcinoma arising from an area where shallow ulceration preceded tends to assume a more anaplastic histological pattern,

representing itself as a scirrhus carcinoma in advanced stage of infiltration.

The findings in the present study on the more advanced cases of scirrhus cancer are apparently in agreement with the assumption above. Inability to produce the direct evidences of precedent ulcers in the fields of cancerous infiltration in the rest of the material is no wonder, since destruction of such evidences by the advancing cancers is quite natural.

We may add another very suggestive finding as to the histogenesis of scirrhus carcinoma. One of the cases reported by Asakura in his series of multicentric malignancies of the stomach revealed two independent foci of scirrhus carcinoma. Both were quite small and had occurred in association with chronic ulcers, UL-II and UL-III respectively. Four of other double cancer cases, had a medullary carcinoma and a scirrhus carcinoma each. The latter was limited in size and invariably associated with a chronic ulcer.

Thus, it may not be too far going to assume that a scirrhus carcinoma of the stomach arises from the area where a chronic ulcer has been present before. The vice-versa is not true, however: there are many evidences that medullary varieties of carcinoma may originate from the edge of a chronic ulceration. An entire set of conditions which should regulate the formation of the scirrhus carcinoma in the stomach is still unknown. It has become clear that precedent chronic ulcers, especially the shallower ones, at least play a major role. Further more, it is suggestive that, if the gastric mucosa at the regenerating margin of an ulcer is of the metaplastic variety, the cancer arising in that area is more likely to be growing medullary and not scirrhus. The intestinal-metaplastic epithelium of the stomach tends to give rise to a more well differentiated type of carcinoma.

It is important to point out that, if all the scirrhus carcinomas of the stomach arise from previous ulcerative lesions, more than 40% of all gastric cancers among the Japanese should have benign chronic ulcers as their morphological precursors. There is a statistical parallelism between the incidences of gastric cancer deaths and deaths due to gastroduodenal ulcers among many nations. The Japanese maintains top-levels in the both death causes. If there are many deaths due to ulceration there should be much more cases which have shallower ulcers not leading to immediate death.

Table 5. Sex and Age Distribution of Scirrhus Carcinomas of the Stomach

	-19	20-29	30-39	40-49	50-59	60-69	70-79	Total
Male	0	5	24	66	66	51	7	219
Female	1	10	35	60	70	26	3	205
Total	1	15	59	126	136	77	10	424

Table 6. Sex and Age Distribution of Scirrhus Carcinoma of the
Stomach: % of Scirrhus in all Carcinomas

* All carcinomas 1793 ** scirrhus 424

Age	10-19	20-29	30-39	40-49	50-59	60-69	70-79	80-89
Male	—	38.4%	46.6%	31.5%	21.9%	15.9%	14.2%	0%
Female	100%	84.6%	60.9%	48.4%	31.3%	25.3%	25.0%	0%
M+F	100%	61.1%	53.2%	38.2%	24.9%	18.6%	18.5%	0%

CONCLUSION

1. On the basis of histologic examination of a large series of carcinomas of the stomach, histological precursors of scirrhus variety of cancer is discussed.
2. It is revealed that scirrhus carcinoma mostly originates in association with preexisting benign ulcerative lesion of the stomach.
3. It was emphasized that among the ulcerative changes shallower lesions predominate in the causation of scirrhus carcinoma.
4. This assumption leads to a more important conclusion that gastric cancer among the Japanese originates very frequently from the benign ulcer.

日本癌学会会員名簿

(1961 年 4 月 1 日 現在)

役

員

会 長 黒川利雄

幹 事

赤崎兼義	今井環	岸三二	黒川利雄	久留勝	中原和郎
太田邦夫	武田勝男	滝沢延次郎	田崎勇三	八木日出雄	吉田富三

評 議 員

赤崎兼義	天野重安	荒木千里	綾部正大	藤井隆	藤森正雄
藤浪得二	福岡文子	浜崎幸雄	日比野進	樋口一成	平木潔
今井環	今永一	入江英雄	石館守三	岩鶴竜三	陣内伝之助
梶原 彊	梶谷 鑠	釜洞醇太郎	加来道隆	桂重次	川俣順一
岸三二	小林隆	黒川利雄	久留勝	楠隆光	牧野佐二郎
増淵一正	三谷靖	宮地徹	三宅仁	水野伝一	森和雄
村上忠重	長与健夫	中原和郎	中山恒明	櫛林和之	落合京一郎
岡本肇	太田邦夫	堺哲郎	桜井欽夫	佐藤八郎	佐藤春郎
瀬木三雄	島田信勝	新保幸太郎	白羽弥右衛門	須田正巳	鈴江懐
武田勝男	武内忠男	滝沢延次郎	田内久	田崎勇三	友田正信
塚本憲甫	卜部美代志	白淵勇	和田武雄	八木日出雄	山形敏一
山口寿	山本正	山下久雄	吉田富三		

名 誉 会 員

布施信良	大阪市東区法円坂町	国立大阪病院	中川 諭	札幌市大通西 26 丁目
今村荒男	西宮市南郷町 57		緒方 知三郎	東京都文京区駒込曙町 23
勝沼精蔵	名古屋市東区七小町 4		大島 福造	名古屋市昭和区御器所町 1 の 10
木村哲二	東京都港区赤坂青山 5 の 36		Prof. Hans. von Euler	
木下良順	Medical Research Institute		Biochemical Institute, Stockholm	
	City of Hope, 1500 E. Duarte		University, Stockholm, Sweden	
	Road, Duarte, California, U.S.A.		佐々木 隆興	東京都千代田区神田駿河台 2 の 2
松本信一	京都市左京区浄土寺西田町 61		沢田 藤一郎	福岡市大濠 144
森茂樹	京都市左京区岡崎真如堂前 17		塩田 広重	東京都文京区本郷弓町 1 の 10

杉浦 兼松 Sloan-Kettering Institute for
Cancer Research, 444 East 68th
Street New York 21, N.Y.,

U.S.A.

津田 誠次 岡山市広瀬町 187

一 般 会 員

A

阿波 章夫 札幌市 北大理学部動物学
安倍 弘昌 市川市国府台 国立国府台病院
阿武 保郎 米子市鳥取大放射線科
足立レオナルド Department of Medicine Hos-
pital del Empleado
相羽 達雄 東京都文京区本富士町 東大医学部
木本外科
相沢 憲 東京都板橋区大谷口町 日本大学医
学部細菌学教室
明石 章 豊中市蛸ヶ池 阪大薬学部薬物学教
室
明石 勝英 札幌市 札幌医大産婦人科
赤井 貞彦 新潟市水道町 1
赤堀 四郎 大阪市北区中之島 阪大理学部
赤木 正信 熊本市北新坪井町 228 柏木方
赤木 制二 岡山市岡 164 岡山大学医学部附属
病院第二外科
赤倉 一郎 東京都新宿区信濃町 慶大病院外科
赤嶺 俊 東京都板橋区板橋 日大病院若林外
科
赤崎 兼義 仙台市 東北大学医学部病理
赤塚 謙一 東京都中央区日本橋本町 3 の 3 中
外製薬学術部
秋吉 正豊 東京都文京区湯島 3 の 1 東京医科
歯科大学病理学教室
青木 みか 名古屋市瑞穂区汐路町 3
青木 重久 京都市左京区 京大医学部病理
青木 孝好 東京都大田区古市町 279
青木 忠 広島市 広島大学医学部皮膚泌尿器科
青木 忠夫 逗子市逗子 867
青木 行俊 大阪市福島区堂島浜通 大阪大学医
学部附属病院第二外科
青山 敬二 大阪市福島区海老江上 1 の 57
青山 撰 大阪市南区内安堂町通 1 の 28
青柳 安誠 京都市 京都大学医学部内科
青柳 昌樹 東京都港区芝愛宕町 慈恵医大産婦
人科
天野 重安 京都市左京区吉田 京都大学ウィル

ス研究所

天野 武彦 京都市北区小山上内河原町 19
網野 三郎 東京都新宿区柏木 1 丁目 東電医大
放射線科
麻田 栄 高槻市古曽部 大阪医科大学麻田内
科
朝倉 元晴 東京都中央区銀座東 癌研病院
朝倉 志郎 高岡市横田新町 木下方
朝比奈 勝 大阪市西淀川区姫島町 1768
浅野 正英 東京都港区白金台町 国立予防衛生
研究所病理
浅野 健夫 岡山市岡 岡山大学医学部平木内科
浅野 定 京都市北区小山下内河原町 80
芦川 和高 東京都品川区大井鮫洲町 205 伊藤
方
熱海 明 山形市香澄町桜小路 山形県衛生研
究所
渥美 理 千葉県船橋市前原公団 57-402
有賀 槐三 東京都杉並区西荻窪 1 の 167
有広 忠雅 東京都港区芝愛宕町 慈恵医大産婦
人科
有留 照周 米子市鳥取大第一病理
東 昭哉 京都市京府医大外科
東 弘 大阪市 阪大第二外科
東 陽一 八幡市黒崎岸の浦 厚生年金病院
新井 志郎 西宮市神楽町 29
新井 正 習志野市大久保町 千葉大学腐敗研
究所
新井 恒人 和歌山市美園町 和歌山県立医科大
学病理
荒木 千里 京都市左京区 京大病院外科
荒木 正哉 京都市上京区河原町広小路 京都府
立大学病理
荒木 嘉隆 東京都文京区本富士町 東京大学医
学部田坂内科
荒毛 正興 熊本市本庄町 483
荒尾 竜喜 熊本市本庄町 熊本大学医学部皮膚科
荒川 順正 東京都品川区 三共研究部
栗屋 博信 宇部市 山口医大病理
綾部 正大 米子市西町 鳥取大学医学部外科

安 齋 哲 郎 札幌市南1条西17丁目 札幌医大
内科

安燈 権八郎 大阪市東住吉区田辺東之町 5~10

安 藤 盛 夫 東京都板橋区大谷口町 日本大学医
学部比企内科

安 藤 隆 東京都板橋区 日大病院第一外科

B

馬 場 為 義 大阪市阿倍野区旭町 大阪市立大学
医学部病理

馬 場 恒 男 東京都豊島区西巢鴨2 癌研究所内

戸 次 英 一 札幌市南一条西 札幌医大内科

C

千 葉 文 雄 仙台市東2番丁 129 仙台通信病院

千 葉 胤 孝 名古屋市中区七小町 61

千葉 ヨリエ 東京都新宿区市ヶ谷 東京女子医大
産婦人科

地土井 襄重 愛媛県八幡浜市 市立病院

D

土 肥 淳 二 大阪市北区堂島西町 3 阪大微研外
科

土 井 一 雄 大阪市東区今橋3 湯川胃腸病院

土 井 修 大阪市阿倍野区阪南町東4の26

土 手 剛 東京都新宿区東大保 東京医科大学
病理

E

蛭 名 勝 昭 土浦市 3550 新治協同病院

海老名 敏明 仙台市 東北大学抗酸菌病研究所

遠 藤 英 夫 仙台市北四番丁 62 東北大学抗酸
菌病研究所

遠 藤 英 也 東京都豊島区西巢鴨2 癌研究所内

遠 藤 浩 良 東京都文京区本富士町 東大薬学生
理化学

遠 藤 三 郎 新潟市旭町 新潟大学医学部外科

遠藤 辰一郎 福島市杉妻町 福島医大遠藤外科

遠城寺 宗知 福岡市草芥江本町 3の89

榎 本 宏 東京都文京区 東大薬学部生化学

榎 本 金 吾 埼玉県北足立郡戸田町下戸田 田辺
製薬東京研究所

榎 本 真 東京都文京区東大病理

江 頭 清 之 東京都港区芝白金台町 国立予防研
病理部

F

淵 上 在 弥 東京都中央区銀座東癌研究所病院

藤 井 源七郎 東京都港区芝白金台町 伝研附属病
院外科

藤 井 純 一 長崎市坂本町 93 長崎大学附属病
院産婦人科

藤 井 信 之 東京都板橋区大谷口町 日大医学部
比企内科

藤 井 隆 東京都文京区 東京大学理学部動物
学教室

藤 間 利 行 熊谷市筑波町 904

藤 巻 雅 夫 新潟市旭町 新潟大学医学部外科

藤 森 明 良 岡山市岡 岡山大学医学部平木内科

藤 森 速 水 大阪市阿倍野区旭町 大阪市立大学
医学部産婦人科

藤 森 正 雄 東京都千代田区神田和泉町 1 三井
厚生病院

藤 本 和 生 熊本市本荘町 熊本大学医学部第二外
科

藤 本 慶 治 東京都千代田区神田美土代町 島津
製作所

藤 浪 修 一 名古屋市名市大外科

藤 浪 得 二 大阪市福島区堂島浜通3丁目 大阪
大学医学部皮膚科教室

藤 田 順 一 東京都港区芝伊皿子 17

藤 田 承 吉 東京都大田区調布嶺町 1の99

藤田 吉四郎 東京都文京区 東大医学部木本外科

藤 咲 暹 仙台市 東北大学医学部公衆衛生学
教室

藤 沢 圭 吾 大阪市東区道修町 塩野義製薬

藤 沢 正 男 大阪市東淀川区加島町 田辺製薬株
式会社

藤 沢 俊 郎 東京都小金井市貫井 957 藤沢薬品
東京研究所

藤 高 道 也 広島市大手町 7の10

藤 原 二 朗 大阪市阿倍野区文の里 2の48

藤 原 順 神戸市生田区楠町 神戸医大第一内
科

藤 原 剛 宇部市小津 山口医科大 第二外科

福 地 竜 夫 熊本市本荘町 熊本大学医学部第二外
科

福地 言一郎 東京都中央区銀座2の1 三共製薬
企画部

福 田 昭 吉 大阪市福島区 阪大医学部武田外科

福 田 勝 次 高槻市古曽部 大阪医科大学麻田外

科
 福田 保 東京都文京区 順天堂大外科
 福井 謙一 京都市左京区吉田本町 京都大学工
 学部燃料化学教室
 福井 繁三郎 大阪市北区堂島西町 大阪市外電話
 局保健課
 福井 享 大阪府吹田市砂子町1の2790
 福西 亮 鹿児島市下町 鹿大第一病院
 福岡 文子 東京都豊島区西巢鴨2丁目 癌研究
 所内
 福岡 善見 檀原市畝傍 奈良県立医科大学病理
 福土 主計 仙台市東北大抗研
 福島 正昭 東京都品川区五反田5の55 関東通
 信病院婦人科
 福島 範子 東京都新宿区戸山町1 国立東京第
 一病院病理
 福島 鉄雄 東京都目黒区中目黒3の935の1
 木口方
 福島 清治 弘前市弘大病理
 舟生 一義 東京都杉並区方南町 72
 古江 尚 東京都中央区銀座東 癌研病院
 伏木 信夫 京都市左京区 京大外科

G

源田 哲郎 東京都文京区 日本医大真柄産婦人
 科
 五味 誠 東京都大田区馬込町東4の5
 後藤 裕之 檀原市 奈良医大病理
 後藤 俊 岐阜市司町 40 岐阜県立医大第一
 内科

H

芳賀 圭五 愛知県春日井市高蔵寺町 名古屋第
 一日赤
 萩原 昭夫 豊中市北刀根山 阪大薬学
 萩原 忠文 東京都板橋区大谷口町 724 日大医
 学部比企内科
 箱守 仙一郎 仙台市 東北薬大癌研
 浜崎 幸雄 岡山市 岡山大学医学部病理学教室
 浜崎 靖 大阪市福島区 阪大病院放射線科
 浜崎 美景 岡山市岡 岡山大学医学部病理
 宇部市 山口医大病理
 浜田 忠雄 福岡市堅粕 九州大学医学部第一内
 科血液研究室
 花田 昭 弘前市 弘前大学医学部第一外科
 花木 昭 千葉市黒砂町 放射線医学研究所

嘩道 二郎 大阪市東区道修町 武田薬工開発部
 羽野 寿 大阪府豊中市北刀根山 382 大阪大
 学薬学部薬物化学教室
 羽生 文武 大阪市旭区千林町3の249
 原 一夫 大阪市福島区 阪大病院放射線科
 原 義雄 新潟市旭 町新潟大学医学部桂内科
 原 義人 岡山市 岡大生化学
 原見 権一 大阪市阿倍野区旭町 大阪市立大学
 産婦人科
 原田 一洋 宮城県栗原郡若柳町 公立若柳病院
 原田 敬一郎 福岡市堅粕 九大医学部放射線科
 原田 稔 京都市 京都大第一外科
 原田 幸昭 福岡市九大第二外科
 原田 種一 東京都世田谷区羽根木町 1740
 原田 敏雄 東京都渋谷区大向通 23
 原田 良 札幌市北一条 市立札幌病院内科
 原田 充善 東京都大田区大森 東邦大学病院第
 一内科
 張間 行直 弘前市相良町 弘前大医学部腫外科
 長谷 芳美 山口市後河原 山口大学文理学部生
 物学
 長谷川 圭吾 新潟市旭町 新潟大学医学部外科
 長谷川 正男 仙台市 東北大黒川内科
 長谷川 俊治 名古屋市中区南外堀6の1 国立名
 古屋病院内科
 長谷川 恒範 東京都新宿区東大久保1の 412 東
 京医科大学病理
 橋口 孝雄 大阪府堺市京町通3丁目53の3
 橋口 俊幸 鹿児島市 鹿児島大医学部第一外科
 橋本 清 岡山市岡 岡山大学医学部産婦人科
 橋本 正淑 札幌市 札幌大婦人科
 橋本 嘉幸 東京都足立区四ツ家町 347
 橋本 美智雄 福岡市九大病理
 橋本 修治 鹿児島市薬師町70の2
 橋本 邦久 仙台市 抗研
 橋本 省三 東京都大田区北千束町 455
 橋本 義雄 名古屋市昭和区鶴舞町 名古屋大学
 医学部第一外科教室
 橋爪 藤光 山口県厚狭郡山陽町 国立殖生療養
 所
 蓮田 清 東京都港区芝愛宕町 慈恵医大産婦
 人科
 秦 清三郎 東京都品川区北品川4の78
 秦 藤樹 東京都港区芝白金三光町 138
 畑中 正一 京都市左京区 京大医学部医化学
 波多野 輔久 神戸市生田区楠町神戸医大第二病理

服部 宏己 名古屋市 名大日比野内科
服部 和彦 名古屋市南区柴田本通 4 の13
服部 正次 大阪市福島区堂島浜通 大阪大学医学部堂野前内科
服部 孝雄 東京都品川区五反田 6 の206
林 宏 徳島市蔵本町 1 丁目 県立中央病院外科
林 活次 名古屋市市大病理
林 誠 沼津市 千葉大学腐敗研究所
林 伸夫 神戸市 神戸医大第一病理
林 清四郎 熊本市 熊大薬学部
林 周一 東京都文京区湯島 順天堂医大外科
林 良昭 鹿児島市 鹿児島大学佐藤内科
林 雄俊 高槻市古曽部 大阪医科大学麻田外科
林 裕造 尼ヶ崎市今福 192 塩野義研究所
林 鋭雄 富山県西砺波郡石動町 石動厚生病院
鮎 松 洋 東京都大田区大森 東邦大医学部薬理
日比野 進 名古屋市昭和区 名大医学部内科
東 達郎 鹿児島市山下 鹿児島大第二内科
引地 芳子 仙台市北四番丁 東北大抗研
平福 一郎 東京都品川区五反田 5 の55 関東通信病院臨床検査科
平井 秀松 東京都文京区本富士町 1 東京大学医学部生化学教室
平井 二郎 仙台市 東北大黒川内科
平井 秀 富山市堀川小泉町 806
平井 得夫 長崎市坂本町 長崎大篠島内科
平井 義則 千葉市矢作町 千葉大学医学部病理
平出 光 仙台市 東北大公衆衛生
平木 潔 岡山市 岡大内科
平光 吾一 東京都武蔵野市吉祥寺町 1306
平野 良雄 東京都中央区銀座東 癌研病院
平尾 正 東京都新宿区 慶大外科
平田 良三 金沢市 金沢大学結核研究所化学部
平山 雄 東京都港区芝白金台町 公衆衛生院
広野 巖 名古屋市昭和区鶴舞町 名古屋大学医学部病理学教室
広瀬 文男 広島市霞町 広島大学医学部病理
広瀬 肇 横浜市中区常盤町 5 の68
広瀬 清一 名古屋市瑞穂区 名古屋市立大医学部第一病理
広瀬 俊太 高槻市古曽部府営高槻住宅 31
蛭海 啓行 和歌山市 和歌医大第一解剖

久田 忠男 千葉市稲毛台町 59
菱川 創一 吹田市千里山 312
檜 沢 一夫 徳島市蔵本町 徳島大医学部病理
本郷 敏郎 弘前市相良町 弘前大医学部病理
本郷 基弘 岡山市 岡大婦人科
本間 康正 千葉市玄鼻町 3 の3 千葉大学医学部中山外科
本庄 一夫 金沢市土取場永町 金沢大医学部外科
堀 浩 札幌市北八条 北海道大学理学部動物学教室
堀 宏行 沼津市大久保町 千葉大学腐敗研究所
堀 啓二 大阪市福島区堂島浜通 3 丁目 大阪大学附属病院放射線科
堀江 滋夫 東京都文京区本富士町 1 東京大学生化学
堀井 秀夫 名古屋市北区鳩岡町 1 の1 志賀住宅 35 棟 407
堀江 健也 東京都北区滝の川 6 の33
星 川 信 名古屋市中区新栄町 3 の29 名古屋大学分院外科
星野 章 名古屋市昭和区鶴舞町 名古屋大学医学部第一内科
星野 寿雄 東京都大田区池上町 570
星野 宗光 名古屋市千種区鍋屋上野町3281の2 千有荘 140
星野 智雄 東京都中央区銀座東 癌研
細川 修治 宇部市中宇部 山口医大病理
細川 務 東京都港区芝田村町 慈恵医大産婦人科
細田 仁 東京都世田谷区上北沢町 3 の877
細谷 憲政 東京都新宿区東京女子医大生化学

I

井 昭成 熊本市 熊大医学部第二外科
井 洋平 大阪市北区常安町 33 大阪大学医学部第一病理学教室
井 林 淳 札幌市南一条西 17 丁目 札幌医大癌研
市場 邦道 榎原市畝傍 奈良医大病理
一井 昭五 千葉市黒岩町 250 放医研 生理病理部
市川 辰己 茨城県西茨城郡岩間町旭町
市来 輝也 鹿児島市 鹿児島大 佐藤内科
一戸 喜兵衛 札幌市北十四条西五丁目 北大病院

産婦人科

井出 源四郎 千葉市亥鼻町 千葉大学医学部病理

飯 島 登 東京都目黒区白金台町 東大伝研

飯 田 正 章 名古屋市昭和区鶴舞町 名古屋大学
医学部産婦人科教室

猪 狩 定 典 東京都豊島区西巢鴨 癌研

井口 登美子 東京都新宿区市ヶ谷 東京女子医大
病院産婦人科

池 田 作 哉 北海道芦別市旭町油谷 1 油谷芦別
炭礦病院

池 田 精 孝 東京都調布市下石原 2524

池 田 昭 二 熊本市出水町国府 823

池 尻 泰 二 福岡市堅粕 九州大医学部第二外科

池 内 彦 名古屋市瑞穂区 名古屋市立大医学
部病理

稲 葉 午 郎 静岡県藤枝市前島 立市志太病院

稲 津 佳 彦 東京都品川区西品川 1 の 888 三共
株式会社研究部

井 上 一 男 宇部市中宇部 山口医大病理

井 上 正 東京都新宿区信濃町 慶応外科

井 上 正 順 大阪市福島区 阪大癌研究所

井 上 諒 京都市左京区浄土寺下南田町 9

井 上 武 彦 大阪市東住吉町西今川町 6 の 28

乾 朝 郎 和歌山市 和歌山医大耳鼻科

乾 直 道 徳島市蔵本町 徳島大医学部病理

乾 成 美 岐阜市司町 40 岐阜医大第一内科

今 井 大 仙台市北四番丁 東北大医学部病理

今 井 弘 子 東京都北区西ヶ原 1 の 26 薬理研究
所

今 井 環 福岡市箱崎町 九大医学部病理

今 村 博 東京都北区西ヶ原町 1 の 26 薬理研
究所

今 村 弘 熊本市本荘町 483 熊本大学病院産
婦人科

今 永 一 名古屋市昭和区鶴舞町 65 名古屋
大学附属病院外科

今 西 幸 雄 大阪市旭区赤川町 2 の 43

今 岡 健 郎 弘前市相良町 弘前大学医学部第一
外科

五百蔵 昭夫 神戸市生田区楠町 神戸医大第一外
科

入 江 英 雄 福岡市九大放射線科

入 野 昭 三 岡山市岡 岡大医学部平木内科

石 原 実 名古屋市昭和区鶴舞町 55 名古屋
大医学部産婦人科

石 原 隆 昭 甲府市国玉町 1018

石 橋 梯子 東京都板橋区 日大細菌学教室

石 部 知行 広島県安芸郡海田市町 2096

石 橋 昭 東京都板橋区大谷口町 日本大学医
学部細菌教室

石橋 嘉久蔵 八戸市大字番丁 44 石橋内科

石 橋 幸 雄 東京都港区芝白金台町 1 の 39 伝研
外科

石 田 健 蔵 大阪市北区常安町 大阪大学医学部
第一病理

石田 名香雄 仙台市 東北大医学部細菌学

石 田 哲 哉 神戸市生田区楠町 神戸医大放射線
科

石 館 守 三 東京都文京区 東京大学薬学部薬品
分析化学教室

石 館 基 東京都杉並区高円寺 4 の 608

石 館 卓 三 仙台市 東北大病理

石 上 重 行 大阪府北河内郡門真町門真 58

石 黒 熊 夫 大阪市東区道修町 2 の 50 稲畑産業
KK 医薬品部

石 井 良 治 東京都渋谷区原宿 2 の 170

石 井 暢 東京都新宿区戸山町 国立東一病院
検査科

石井 善一郎 松本市旭町 信州大医学部病理

石 川 育 夫 大阪府大東市寺川 71 の 1

石 川 二 郎 大阪市天王寺区筆ヶ崎町 50 大阪
赤十字病院研究科

石 川 正 臣 東京都文京区西片町 10 いの 1

石 川 通 夫 名古屋市中区新栄町 3 の 29 名古屋
大学医学部分院外科

石 川 七 郎 東京都新宿区信濃町 慶応病院

石川 大刀雄 金沢市 金沢大学医学部第二病理

石 河 利 隆 東京都品川区五反田 5 の 55 関東通
信病院

石 木 哲 夫 東京都青梅市西分 298 青梅市立総
合病理検査室

石 倉 肇 北海道岩内町大和 115

石 岡 国 春 仙台市北四番丁 東北大学医学部黒
川内科

石 山 秀 彦 東京都新宿区信濃町 慶応外科

石 山 俊 次 東京都品川区五反田 5 の 55 関東通
信病院外科

石 崎 昭 一 新潟市旭町 新潟大学医学部外科

石 沢 敏 子 東京都千代田区神田駿河台 2 の 2
佐々木研究所

石 塚 隆 和 東京都北多摩郡狹江町 慈恵医大第
三病院内科

磯 貝 豊 千葉市亥鼻町 千葉大医学部歯科口腔外科
磯 橋 保 高槻市古曽部 大阪医科大麻田外科
磯 島 晋三 大阪市北区 阪大医学部産婦人科
磯 松 俊夫 札幌市 北大医学部第二外科
伊 丹 康人 東京都港区芝田村町 慈恵医大病院整形外科
伊 藤 剛二 広島市皆実町 3 の103の18
伊 藤 治 英 東京都港区芝愛宕町 慈恵産婦人科
伊 藤 英太郎 大阪福島区堂島浜通 阪大癌研
伊 藤 一二 徳島市寺島本町西 3 徳島市民病院
伊 藤 薫 東京都港区芝白金台町 伝研細胞化学
伊 藤 正 敏 仙台市 東北大医学部黒川内科
伊 藤 信義 神戸市生田区楠町 神戸大第一外科
伊 東 信 行 橿原市畝傍 奈良県立医大病理
伊 藤 亨 岐阜市司町 岐阜医大乾内科
伊 藤 辰 治 新潟市旭町通一番町 大学本部
伊 藤 哲 夫 札幌市北十二条西五丁目 北海道大学医学部第一病理
伊 藤 敏 夫 千葉市 千葉大医学部中山外科
伊 藤 嗣 郎 静岡市梅屋町 1 の 7
伊 藤 安 彦 仙台市 東北大抗酸菌研究所
糸 井 壮 三 大阪市福島区堂島浜通 大阪大学病院泌尿器科
糸 氏 英 吉 大阪市住吉区中加賀屋町 4 の65 住之江病院
岩 井 正 二 松本市信州大産婦人科
岩 城 徹 大阪市東区道修町 塩野義製薬 KK
岩 木 年 中 大阪市東成区中道本通 1 の12 岩木病院
岩 永 保 人 福岡市 九大病理
岩 崎 一 郎 岡山市内田本町 235
岩 崎 博 長崎市坂本町 93 長崎大病院産婦人科
岩 崎 健 資 熊本県菊地郡西志志村再春荘 官校舎 7
岩 崎 基 高知市 国立高知病院
岩 崎 迪 雄 千葉市 千葉大医学部中山外科
岩 田 平 太 郎 大阪府豊中市北刀根山 382 阪大薬学部薬物学教室
岩 田 克 美 岡山市岡 岡大医学部浜崎病理
岩 田 正 晴 東京都港区芝愛宕町 慈恵医大産婦人科
岩 田 豊 愛知県一宮市富古場町 2
岩 鶴 竜 三 和歌山市和歌浦高松 89

井 坂 英 彦 東京都千代田区神田駿河台 2 の 2 佐々木研究所
井 坂 英 雄 津山市 作陽短期大学
伊 沢 隆 太 東京都大田区大森 東邦大医学部薬理
伊 沢 立 身 久留米市 久留米大耳鼻咽喉科
井 添 五 郎 長崎市坂本町 長大医学部産婦人科
泉 彪之助 金沢市土取場町 金沢大医学部病理
和 泉 昇 次 郎 秋田県本荘市 由利組合総合病院
泉 洋 神戸市生田区 神戸医大第二外科
泉 雄 勝 東京都千代田区神田和泉町 1 三井厚生病院外科

J

自 見 昭 司 長崎市坂本町 長崎大医学部産婦人科
陣 維 喜 東京都渋谷区宮代町 日赤中央病院病理
神 保 昭 典 東京都大田区大森 1 の188
仁 礼 裕 神戸市生田区 神戸大第一外科
陣 内 伝之助 岡山市 岡山大第一外科
陣 立 恒 夫 日立市河原字町 1883

K

門 根 謙 介 豊中市旭丘団地 43 の 402
香 川 恒 雄 静岡県駿東郡長泉村下土狩 1188 協和醸酵富士工場
香 川 靖 雄 東京都文京区東大医学部生化学
梶 英 雄 東京都目黒区三谷町 29
可 見 三 郎 豊中市北刀根山 382 大阪大学薬学部薬物学教室
梶 原 疆 大阪市東淀川区十三西之町 4 の54 武田薬品工業研究所
梶 谷 銀 東京都中央区銀座東 癌研病院
梶 谷 清 己 岡山県邑久郡久町立病院
笈 弘 毅 千葉市 千葉医大放射線
笈 守 京都市上京区元誓願寺通大宮東入山根方
垣 内 洋 二 横浜市戸塚区原宿 国立横浜病院松月寮
角 谷 哲 司 広島市 広島大医学部産婦人科
釜 洞 醇 太 郎 大阪市東住吉区山坂町 3 の37
鎌 田 忠 夫 千葉市 千葉大中山外科
亀 井 秀 雄 名古屋市中区 名大分院外科
亀 田 幸 雄 金沢市 金沢大学薬学部
亀 谷 哲 治 仙台市北四番丁 東北大医学部薬学

科薬品製造教室

亀山 英之 岡山市 岡山大陣内外科
 神前 五郎 豊中市刀根山2の171
 神前 武和 京都市北区小山板倉町3
 紙野 建人 大阪市西成区辰巳通2の14 松田病院
 神原 信明 大阪市北区 阪大医学部病理
 神原 武 熊本市本荘町 熊本大学医学部病理学教室
 神崎 一吉 仙台市小田原長丁通1
 神田 昇 一宮市明神津18
 神田 昌一 東京都文京区 東大理学部生化学寺山研究室
 金淵 一郎 仙台市 東北大抗研
 金久 武晴 神戸市御影局 神戸大理学部
 金子 仁 東京都新宿区戸山町 国立東一病院病理
 金子 君枝 東京都港区芝白金台町 伝研組織培養室
 金木 悟 東京都足立区柳原町150
 蟹沢 成好 千葉市亥鼻町 千葉大医学部病理
 加来 道隆 熊本市大江町本543 熊本大医学部産婦人科
 笠原 靖史 豊中市本町1の18
 笠井 沖 北海道空知郡富良野町 富良野協成病院
 笠松 達弘 東京都文京区本富士町 東大医学部産婦人科
 柏井 浩三 大阪市 阪大病院泌尿器科
 春日 孟 東京都豊島区西巣鴨2丁目 癌研究所内
 方波見 康雄 北海道空知郡奈井江町
 片桐 謙 尼崎市今福192 塩野義製薬研究所
 片桐 昭文 新潟市新潟大第一外科
 片山 正治 西宮市染殿町60 西宮市立市民病院
 加藤 勲 東京都大田区調布千鳥町59
 加藤 出 東京都渋谷区千駄ヶ谷4の13
 加藤 俊 東京都港区芝愛宕町 慈恵医大産婦人科
 加藤 辰吉 東京都文京区駒込日本医大病院 真柄産婦人科
 加藤 篤二 広島市 広大皮膚科
 加藤 次男 大阪市北区常安町33 大阪大学医学部第一病理
 加藤 義昭 名古屋市中千区元古井町6の3
 勝久 文雄 熊本市本荘町 熊本大学第二外科

勝井 富三郎 大阪市福島区 阪大医学部附属病院武田外科
 桂 重次 仙台市北四番丁85 東北大学医学部外科
 桂 佐元 盛岡市 岩手医大病理
 勝田 甫 東京都港区芝白金台町 伝研病理部
 勝屋 弘辰 熊本市本荘町 熊本大学第二外科
 賀内 俊介 兵庫県小野市大島町1094
 川端 和利 札幌市南5条西6丁目
 川田 光三 東京都新宿区 慶応大外科
 川口 一江 東京都港区芝白金台町 東大伝研組織培養
 川平 重雄 鹿児島県加世田市武田5545 米川方
 河合 和夫 津市島居町 三重県立大医学部病理
 河合 直次 千葉市葛城町
 河合 信秀 東京都中野区昭和通2の29
 川井 一男 布施市永和3丁目74の7
 川井 潔 岡山市岡 岡大医学部附属病院平木内科
 川上 博 東京都新宿区市ヶ谷 東京女子医大病院産婦人科
 川岸 一郎 金沢市 金沢大医学部第二外科
 川俣 順一 大阪市北区堂島町 大阪大学微生物研究所
 川俣 建二 東京都文京区湯島順天堂外科
 川守田 勇一 北海道芦別市本町649
 川村 謙二 京都市上京区京都医大 川村外科
 川村 弘徳 東京都杉並区上荻窪2の227 野村方
 川村 範夫 岡山市 岡山大学平木内科
 川村 太郎 東京都文京区 東大皮膚科
 河村 虎太郎 広島市大手町1の33
 川村 祐二 和歌山市海草郡下津町大字下津768
 川名 衛 東京都港区芝白金台町 東大伝研組織培養
 川名 正直 千葉市 千葉大医学部中山外科
 河西 孝信 広島市翠町 広島県立病院産婦人科
 河野 極 徳島市蔵本町 徳島大医学部第一外科
 河崎 明彦 東京都中野区氷川町25
 川崎 明徳 岡山市西中山下89
 川崎 裕也 仙台市北四番丁 東北大学医学部医化学教室
 川崎 祐宣 岡山市西中山下89
 川崎 雅康 東京都新宿区 慶応病院
 河瀬又右衛門 貝塚市木積880

河 瀬 収	熊本市城内二の丸 熊本大学体質医学研究所病理	北 村 四 郎	新潟市 新潟大医学病理
川 島 吉 良	名古屋市昭和区鶴舞町 名古屋大学医学部産婦人科	北 村 旦	橿原市奈良医大病理
河 津 竜 介	熊本市本荘町 483 熊本大学病院産婦人科	北 野 邦 俊	熊本市本荘町 熊本医学部第二外科
河 内 実 世	宮崎市高千穂通 県立宮崎病院病理	北 沢 英 雄	東京都渋谷区上通 2 の 27 北沢胃腸医院
河内 健次郎	東京都中央区日本橋 塩野義製薬 K K	喜多島 康一	岡山市岡 岡大医学部平木内科
香 山 時 彦	和歌山市 和歌山大学芸学部生物	木 山 敏	岡山市弓之町 78 白神方
川 添 隆 茂	熊本市新細工町 56 本田方	清 野 祐 彦	山形市東原町 360
香 月 秀 雄	千葉県矢作町 千葉大医学部河合外科	小 林 昭 夫	東京都北区袋町 2 の 1470
木 田 威 俊	金沢市上弓の町 19 の 5	小 林 忠 義	東京都新宿区信濃町 慶大医学部病理
木 畑 正 義	岡山市岡 岡大平木内科	小 林 春 満	福岡市 九大外科
貴 家 寛 而	福島区杉妻町 福島医大産婦人科	小 林 久 人	名古屋市昭和区 名大第二病理
菊 地 浩 吉	札幌市 北大医学部第一病院	小 林 淳 一	岡山市岡 岡大医学部陣内外科
木 村 郁 夫	橿原市四条町 奈良医大病理	小 林 延 年	千葉市 千葉大医学部病理
木 元 正 二	氷見市鞍川 氷見厚生病院	小 林 節 昭	熊本市京町 2 の 169
木 村 郁 郎	岡山市岡 岡山大学医学部平木内科	小 林 定 道	札幌市 北大医学部第二外科
木村 喜代次	名古屋市鶴舞町 名古屋大学医学部日比野内科	小 林 重 高	東京都港区芝愛宕町 慈恵医大産婦人科
木 村 修 治	神戸市神戸医大放射線科	小林 多喜雄	大阪市北区常安町 阪大微研外科
木 村 正	東京都新宿区戸山町 国立東京第一病院	小 林 隆	東京都文京区本富士町 1 東京大学医学部産婦人学科教室
金 暢 權	東京都北区西ヶ原 1 の 26 薬理研究会研究所 佐藤博方	小 林 輝 夫	東京都港区芝愛宕町 慈恵医大産婦人科
木 下 悦 之	豊田市小坂本町 1 の 58	小 林 勤	京都市北区衣笠大蔵町 36 田中弥郎方
木 下 文 雄	東京都新宿区西大久保 都立大久保病院	香 江 進	福岡市東唐人町 40
衣 川 湍 水	千葉市 千葉大衛生学	古 賀 慶 八 郎	福岡市九大産婦人科
岸 恭 也	徳島市蔵本町 徳島大医学部病理	古 賀 元 晃	福岡市湯洲町 58
岸 三 二	東京都港区広尾町 17	古 閑 睦 好	熊本市二の丸 熊本医大医学部生理
岸 忠 生	熊本市 熊大第二外科	古 賀 良 彦	仙台市 東北大医学部放射線
貴 島 幸 彦	大阪市 阪大医学部癌研	古 閑 義 之	東京都港区芝愛宕町 慈恵医大病院古閑内科
貴 島 亨	鹿児島市 鹿児島大佐藤内科	小 暮 巽	東京都新宿区 慶応大整形外科
岸 本 英 正	名古屋市昭和区鶴舞町 名古屋大学医学部病理学教室	小早川 庸造	東京都千代田区神田駿河台 日本大学歯学部病理
岸 野 泰 雄	徳島市蔵本町 徳島大医学部第一外科	幸 保 文 治	東京都板橋区 日大細菌学
北 畠 隆	名古屋市 名大医学部放射線	小 泉 昭 雄	仙台市 東北大武藤外科
北 出 文 男	高槻市古曽部 大阪医大麻田外科	小 泉 博	土浦市下高津 国立霞ヶ浦病院
北 川 司 良	京都市左京区下鴨岸本町 59	駒 越 喬 貞	名古屋市中区七本松町 2 の 28
喜多村 勇	岡山市岡 岡山大学医学部小児科	小 松 信 彦	東京都港区白金台町 東大伝研
北 村 元 男	岡山市岡 岡大医学部陣内外科	小 森 昭	札幌市 札幌医大産婦人科
北 村 三 郎	東京都港区青山高樹町 15	小 村 裕	金沢市 金沢大学第二内科
		小 村 孝	札幌市北 14 条西 5 丁目 北大病院産婦人科
		今 功	仙台市北四番丁 東北大医学部病理

- 近藤 忠雄 札幌市 北大医学部第1内科
近藤 達平 名古屋市千種区朝岡町 3 の38
近藤 哲 東京都品川区五反田 関東通信病院
産婦人科
近藤 敏 仙台市北四番丁 東北大抗酸菌病理
研究所
近藤 義雄 大阪市西区立売堀南通 日生病院
小西 義男 長野市三輪田町 1 の313 石川外科
病院
康野 明 東京都品川区平塚 6 丁目 昭和医大
医動物
河野 泰道 大阪市北区 阪大微研病院
紺谷 日出雄 大阪市 阪大微研外科
小関 哲夫 弘前市 弘前大病理
越 哲也 和歌山市 和歌山日赤産婦人科
小坂 順造 静岡市小島 静岡薬科大学
香坂 三男 札幌市南一条 札幌医大産婦人科
小関 弥平 札幌市南1条西 17 札幌医大病理
小島 瑞 仙台市北四番丁 東北大医学部病理
小島 清秀 名古屋昭和区鶴舞町 名古屋大学医
学部第二病理
小島 国次 福島市杉妻町 福島医大病理
小島 修 熊本市熊本大産婦人科
小島 昭三 米子市鳥取大桑原外科
小島 庸一 京都市左京区岡崎黒谷町 33 井上
越村 三郎 金沢市土取場永町 15 金沢大学結
核研究所
腰塚 為久 東京都文京区本富士町 東大病院木
本外科
越浦 良三 金沢市 金沢大学結核研究所
小谷 利一 岡山県和気郡大字田原下
小谷 勉 大阪市阿倍野区阪大 整形外科
小塚 亮 岡山市岡 岡大医学部平木内科
小塚 貞雄 名古屋市中区昭和区 名大第二病理
小山 真 新潟市 新潟大第一外科
小山 善之 東京都千代田区五番町 10 の2
久保 久光 東京都中央区銀座東 癌研病院
久保 久雄 和歌山県伊都郡高野口町田原
久保 勝彦 京都市左京区聖護院川原町 京都大
学病院内科第一講座
久保田 盛志 東京都新宿区戸山町 国立第一病院
外科
久保田 富也 弘前市 弘前大松永内科
工藤 祐三 盛岡市内丸 岩手医大第一内科
空閑 秀邦 宇部市東区常盤通 3 丁目 山口県立
医大第一外科
隈部 寿一 熊本県玉名市中 33
雲出 正 仙台市 東北大黒川内科
熊木 敏郎 東京都文京区駒込千駄木町 59 日
本医大栄養学教室
熊本 寛格 岡山市 岡山産婦人科
熊本 享 弘前市 弘前大病理
熊谷 勝男 仙台市 東北大細菌学
熊谷 謙三郎 大阪市天王寺区筆ヶ崎町 桃山病院
久野 敦二郎 東京都中央区銀座東 2 の17 癌研病
院
倉堀 知弘 大阪市北区堂島西町 3 大阪大学微
生物病研究所病院
倉光 一郎 東京都世田谷区祖師谷 2 の41 D739
倉科 達也 土浦市高津町 国立霞ヶ浦病院官
舎
倉田 当助 東京都千代田区神田駿河台 2 の2
佐々木研究所
倉恒 国徳 福岡市堅粕 九大医学部衛生学教室
栗林 宣雄 東京都渋谷区宮代町 日赤中央病院
病理
栗原 登 仙台市 東北大公衆衛生
栗野 玄佐武 福島市杉妻町 福島県立医大第一内
科
栗田 宗次 名古屋市昭和区鶴舞町 名古屋大学
医学部第一内科
黒岩 耕 佐賀県東松浦郡相知町伊岐佐甲 775
玄岡 末雄 西宮市上甲東園 2 の70
黒川 清之 国立市国立町 200
黒川 利雄 仙台市外記丁 10
黒田 行昭 大阪市北区常安町 33 大阪大医学
部遺伝学
黒田 吉男 福岡市堅粕 九州大学医学部医化学
教室
畔柳 繁 東京都中央区銀座東 癌研病院
久留 勝 大阪市福島区 阪大医学部外科
日下部 博 大阪市福島区堂島浜通り 大阪大学
久留外科
草間 悟 茨城県北海道市宝町
久島 環哉 大阪市西淀川区大和田町 600
楠原 富弘 岡山市 岡山砂田外科
楠本 五郎 浦和市太田窪 1461 の2
楠 信雄 広島市霞町 広島大医学部上村外科
楠 隆光 大阪市福島区阪大泌尿器科
桑原 悟 米子市 鳥取大桑原外科
桑野 昭彦 仙台市北四番丁 東北大医学部黒川
内科

桑田 次男 千葉市矢作町 千葉大医学部細菌学教室

京極 方久 京都市左京区 京大医学部病理

M

前田 春雄 福島県勿来市榎田町字本町 63

前田 甚作 滋賀県甲賀郡甲賀町 塩野義油日農場

前田 次郎 和歌山市雅賀屋町

前田 一憲 神戸市生田区 神戸医大放射線

前田 真 和歌山市八番町 7

前田 鹿三 鹿児島市山下町 鹿児島大医学部第2内科

前田 茂和 大阪市阿部野区旭町 大阪市立大学

前田 俊二 鹿児島市 鹿児島大学佐藤内科

前西 浩 神戸市生田区 神戸医大放射線

真柄 正直 東京都文京区駒込千駄木町 日本医大産婦人科

米原 弘 東京都文京区向ヶ丘弥生町 東大応微研

間島 進 仙台市北四番丁 東北大医学部武藤外科

牧野 佐二郎 札幌市北八条 北海道大学理学部動物学教室

真鍋 宮子 神戸市生田区 神戸医大放射線

丸岡 元男 熊本市水前寺本町 174

丸田 正治 鹿児島市山下町 鹿児島大学医学部第二内科

丸山 昌男 橿原市 奈良医大病理

丸山 素弘 東京都品川区西品川 三共 KK 生化学

丸山 孝士 札幌市 北大第一病理

丸山 雄造 松本市旭町 信州大学医学部病理

増淵 一正 東京都中央区銀座東 癌研病院

増田 強三 京都市左京区下鴨芝本町 25

増田 正典 京都市上京区 京都府大内科

益田 忠 熊本市花園町 643 木部方

益山 栄良 東京都文京区本富士町一 東大医学部放射線科

松原 正香 東京都新宿区新小川町2の10 江戸川アパート 65

松田 清 大阪市福島区堂島浜通 大阪大学癌研究所

松田 実 大阪府八尾市佐堂 59

松田 三和 東京都新宿区 慶応大外科

松平 寛通 東京都文京区本富士町一 東京大学

医学部放射線科

松枝 和夫 東京都文京区本富士町 東大医学部産婦人科

松井 英一 東京都北区西ヶ原1の26 薬理研究会研究所

松井 敬介 米子市西町 鳥取大学医学部病理

松井 邦夫 大阪府三島郡三島町大字味舌下 180の66

松 為 実 大阪市北区堂島西町 大阪大学微研病院外科

松井 康能 米子市 鳥取大病理

松前 昭広 東京都港区芝白金三光町 北里研究所

松宮 誠一 東京都千代田区神田三崎町 東京歯大病理

松森 宏 岡山市岡 岡大医学部平木内科

松森 正顕 兵庫県永上郡相原町相原荘

松本 克彦 東京都品川区平塚町六丁目 昭和医科大学医動物学教室

松本 健二 市川市国府台1の2 国立国府台病院放射線科

松本 恭一郎 橿原市 奈良医大病理

松本 光男 東京都文京区 東大理学部生化学

松本 瑞生 神戸市生田区 神戸医大第一外科

松本 哲 熊本市城内二の九町 熊本大学二薬理

松本 真一 京都市中央区西の京門町 38

松本 外史郎 岡山県苫田郡加茂町 第二診療所

松村 悦郎 西宮市丸橋町 119

松村 茂夫 東京都杉並区高円寺7の 960

松村 祐二郎 別府市亀川 国立別府病院研究検査科

松永 藤雄 弘前市相良町 弘前大医学部松永内科

松尾 源一郎 大阪市阿倍野区 阪市大第一外科

松尾 貞雄 和歌山市七番丁1 和歌山医大第一内科

松岡 健司 東京都文京区本富士町 東大医学部清水外科

松岡 規男 東京都中野区新井町2の 461

松岡 雄治 福岡市 九大第二外科

松島 泰次郎 大阪市福島区 阪大医学部癌研

松谷 嘉夫 千葉市栄町 112

松浦 慶元 岡山市岡 岡大医学部微生物

松浦 梅春 岡山市岡 岡山大学医学部砂田外科

松山 研二 群馬県前橋市岩神町 群馬大医学部

	病理		
松山 睦司	名古屋市市中区老松町2の4	高木方	
松崎 功	千葉市 千葉大医学部中山外科		
翠川 宏	名古屋市昭和区 名大第一病理		
南 又一郎	大阪府阿倍野区天王寺町 大阪鉄道 病院外科		
峯 勝	京都市上京区 京府大第一外科		
峰岸 宏年	東京都港区芝愛宕町 慈恵医大産婦 人科		
峰下 鉄雄	吹田市千里山5の1		
水納谷民太郎	福岡市箱崎網屋立筋二		
簀和田 潤	京都市上京区河原町 小路 京都府 立医大荒木病理		
三村 文男	神戸市長田区五番丁8丁目71の2		
三村 久	岡山市 岡山大第一外科		
三上 豊	大阪府貝塚市名越 国立療養所貝塚 千石荘		
三木 吉治	大阪市福島区 阪大皮膚科		
三崎 鈔郎	東京都千代田区神田 東京医大放医 学		
三島 豊	東京都杉並区上高井戸4の1768		
三須 良彦	東京都豊島区西巣鴨2丁目 癌研究 所		
三田村 恭三	東京都港区芝白金台町 伝研組織培 養室		
三谷 茂	東京都渋谷区宮代町一 日赤産院		
三谷 靖	長崎市坂本町 93 長崎医大産婦人 科病理		
水戸 省吾	山形市香澄町 山形県衛生研究所		
三井 文男	東京都大田区大森 東邦大病院第一 内科		
三代 幸彦	東京都千代田区富士見町 日本歯科 大生化学		
宮川 勝馬	甲府市橘町1の79		
宮川 正澄	名古屋市昭和区鶴舞町 名古屋大学 病理		
宮川 正	東京都文京区本郷 東大放射線		
宮木 高明	千葉市矢作町 千葉大薬学部		
三宅 彰	大阪市東淀川区十三西之町 武田研 究所		
三宅 仁	東京都文京区小石川竹早町 82		
三宅 一忠	岡山市岡 岡山大学医学部陣内外科		
三宅 正昭	東京都品川区五反田 関東通信病院 産婦人科		
三宅 毅	岡山市岡 岡山大学平木内科		
宮本 博泰	東京都豊島区西巣鴨 癌研		
宮本 忍	東京都板橋区大谷口上町 日大医学 部外科		
宮村 通敏	長崎市坂本町 93 長崎大学病院産 婦人科		
宮崎 祥子	東京都大田区大森 東邦大学医学部 薬理		
宮崎 秀樹	東京都新宿区柏木 東京医大外科		
宮崎 誠示	金沢市 金沢大第一外科		
宮崎 壮一	兵庫県川西市小花字宮前3の6		
宮崎 吉平	神戸市生田区楠町 神戸医大第二病 理		
宮地 秀樹	大阪市北区常安町 大阪大学医学部 第一病理		
宮地 清一	東京都港区芝赤羽町 3 日本専売公 社東京病院		
宮地 徹	大阪市北区常安町 大阪大学医学部 病理		
宮島 南次	東京都中央区銀座東 癌研病院		
宮脇 英夫	三重県津市広明町 260 竹内伊勢雄 方		
宮沢 政栄	東京都新宿区西大久保1 都立大久 保病院		
三好 秋馬	京都市左京区 京大病院第一内科		
三輪 潔	東京都武蔵野市吉祥寺 1875		
三浦 貴士	倉敷市美和町 1070 倉敷中央病院 放射線科		
三浦 幸二	静岡県浜松市広沢町 123 の1		
三浦 光恵	弘前市 弘大横外科		
三浦 義彰	東京都文京区駒込西片町 10 番地い の1		
溝口 政澄	福岡市堅粕 九州大学医学部第二外 科		
溝口 輝彦	和歌山市 和歌山医大第一内科		
溝越 将城	東京都世田谷区池尻町 9 自衛隊衛 生学校第4科		
御園生 雄三	千葉市 千葉大産婦人科		
溝田 成	東京都葛飾区千葉町 576		
水原 舜爾	岡山市岡 164 岡山大学医学部生 化教室		
水上 哲次	金沢市土取場永町 金沢大学医学部 第一外科		
水野 伝一	東京都品川区上大崎長者丸 284 国 立予防衛生研究所化学部		
水野 潤二	守口市 関西衛大産婦人科		
水野 公明	大阪市東淀川区十三西之町 武田研 究所		

- 水野 通也 大阪市福島区堂島浜通 3 大阪大学
附属病院放射線科
- 水野 忠一 宮城県白石市外北小路 3
- 水本 竜二 金沢市 金沢大本庄外科
- 百瀬 剛一 千葉市矢作町 千葉大医学部 皮膚
泌尿器科
- 百瀬 隆人 東京都豊島区巣鴨 7 の 1696
- 百溪 定七郎 横浜市港北区篠原町 61
- 門前 徹夫 広島市翠町 1751
- 毛利 石子 仙台市 東北大医学部公衆衛生
- 森 一 福島市杉妻町 福島医大中研
- 森 和雄 東京都品川区平塚 6 丁目 昭和医科
大学医動物学教室
- 森 和郷 札幌市 札幌大産婦人科
- 森 昌彦 大阪市北区 阪大歯学部第 2 口腔外
科
- 森 茂美 福岡県嘉穂郡桂川町 明治平山炭鉱
山の手社宅
- 森 淑 福島市杉妻町 福島医大第一内科
- 森 俊一 東京都北区西ヶ原 3 の 22 桑原方
- 森 亘 東京都文京区湯島三丁目 東京医科
歯科大学病理学教室
- 森 芳茂 神戸市生田区楠町 6 丁目 神戸医大
第一病理
- 森口 幸雄 東京都小金井町 2886 桜町病院
- 森口 実 東京都杉並区馬橋 4 の 499 気象研
究所
- 森井 外吉 大阪府枚方市坂 12 関西医大病理
教室
- 森井 健 大阪市北区 阪大微研病院
- 森川 清実 大阪市西区阿波堀通 4 の 20 ガラン
7 病院外科
- 森本 健二 大阪府池田市神田町 1336 公団住
宅 11 の 203
- 森村 義行 大阪市北区常安町 33 大阪大学医
学部第一病理学教室
- 森岡 久 宇部市 東見初病院
- 森岡 哲吾 高槻市古曽部 大阪医科大麻田外科
- 森沢 清 東京都中央区日本橋本町 2 の 5 山
之内製薬 KK
- 森下 和郎 岡山市 岡山大陣内外科
- 森下 宗司 名古屋市昭和区鶴舞町 65 名古屋
大学医学部産婦人科
- 盛田 英明 名古屋市瑞穂区瑞穂通 1 の 27 名古
屋市立大学医学部外科
- 森田 皓三 名古屋市 名大医放線医学
- 森田 澄一 東京都中央区日本橋蠣殻町 4 の 7
- 森田 倫雄 福島市杉妻町 福島医大中央研究所
- 森武 貞 大阪市福島局区内 阪大久留外科
- 森脇 絢子 東京都北区西ヶ原 1 の 26 薬理研究
所
- 森脇 昭介 米子市 鳥取大病理
- 森山 正武 鹿児島市下竜尾町 170
- 守山 隆章 西宮市城山 22 の 1
- 宗像 秀夫 福島市森合字前田 16 の 3
- 棟方 宏次 名古屋市 名大日比野内科第三研究
室
- 村上 晃一 福岡市 九州大学医学部放射線科
- 村上 真 東京都文京区本富士町 東大医学部
産婦人科
- 村上 元正 岡山市岡 岡山大学医学部病理
- 村上 栄 岡山市岡 岡大医学部微生物
- 村上 忠重 東京都港区芝新橋 4 の 30
- 村松 正実 東京都本郷局区内 東大医学部田坂
内科
- 村田 弘行 大阪府守口市南寺方中通 1 の 19
- 村沢 裕啓 東京都武蔵野市吉祥寺 1922
- 村田 栄治 仙台市 東北大黒川内科
- 村田 吉郎 広島市治山 広島 ABCC
- 村田 善保 弘前市本町 弘前大医学部産婦人科
- 村山 浩 千葉市矢作町 千葉大医学部病理
- 村山 博良 東京都杉並区阿佐ヶ谷 4 の 894 田
端方
- 村山 弘泰 東京都中野区宮里町 42
- 室屋 博 東京都中央区日本橋堀留町 1 の 6
台糖ファイザー株式会社
- 牟田 信義 札幌市 札幌医大
- 武藤 幸治 東京都練馬区豊玉上 2 の 2
- 武藤 完雄 仙台市北四番丁 東北大学病院外科
- 武藤 輝一 新潟市旭町 新潟大学医学部外科

N

- 鍋谷 欣市 千葉市支鼻町 千葉大学医学部中山
外科
- 永江 孝直 金沢市泉野町 V17 の 32
- 永末 知 山口市徳山浦石町 徳山中央病院
外科
- 永井 清和 大阪市北区常安町 阪大第二病理教
室
- 永井 清保 堺市大浜南町 230
- 永井 幹男 広島県竹原市忠海町 忠海病院
- 永井 春三 大阪府箕面市半町 171

- 永井 良治 名古屋市昭和区長戸町 3 の33
 永原 貞郎 松本市旭町 信州大学医学部病理学教室
 永堀 善作 浦和市大字大谷口 31 共済会病院
 長石 忠三 京都市左京区 京大結核研究所
 長尾 喜景 東京都千代田区神田 東京歯科大学口腔外科
 長岡 敏雄 東京都大田区大森 東邦大学医学部薬理
 長岡 豊 大阪市福島区 阪大医学部武田外科
 長田 漢珣 東京都大田区大森 東邦大学医学部薬理
 永田 博仁 愛知県豊田市元城町 1 の26
 永田 親義 京都市左京区 京大工学部
 長与 健夫 名古屋市中千種区春里町 1 の24
 内藤 聖二 東京都文京区湯島 順天堂大学第二内科
 中林 登 神戸市東灘区本山町田中 28
 中原 和郎 東京都豊島区目白町 1 の 1141
 中井 育夫 高槻市 大阪医大解剖学
 中居 卓 東京都文京区駒込上富士前町 11
 中泉 正徳 東京都文京区本富士町 東京大学医学部附属病院放射線科教室
 中川 正男 東京都新宿区柏木 1 の 193
 中川 定明 岡山市西中山下 川崎癌研究所
 中川 昌一 札幌市北 14 条西 5 丁目 北大病院第二内科
 中川路 慶一 東京都板橋区大谷口町 日本大学医学部細菌学教室
 中村 章 東京都杉並区方南町 452
 中村 久也 福島市杉妻町 福島医大病理
 中村 博 橿原市四条町 奈良医大病理
 中村 克宏 仙台市 東北大医学部病理
 中村 真 東京都中央区銀座東 癌研病院
 中村 尚孝 札幌市北 12 西 5 北大医学部第二外科
 中村 貞雄 東京都品川区豊町 日本専売公社中央研究所
 中村 史郎 京都市左京区下鴨中川原町 54 吉村方
 中村 義尚 大阪市阿倍野区阪市大第一外科
 中村 嘉三 東京都新宿区信濃町 慶大病院外科
 中村 善次郎 岸和田市西之内町 496 岸和田市立病院
 中野 昭典 岡山市岡 164 岡山大学医学部病理
 中野 勇 東京都中央区日本橋江戸橋 3 の 1
 中野 喜久男 千葉市 千葉大病理
 中野 俊一 大阪市北区堂島西町 3 大阪大学微生物病研究所病院
 中院 邦彦 神戸市生田区加納町 1 の 5 神戸市衛生研究所
 中院 孝円 神戸市灘区 5 毛通 3 丁目
 中島 平太郎 大阪市阿倍野区 阪市大第一病理
 中尾 喜久 前橋市 群馬大学医学部
 中尾 清 福岡市浜の町病院
 中迫 博 名古屋市東区長堀町 3 の13
 中島 哲夫 市川市菅野 東歯大市川病院外科
 中山 広信 弘前市 弘前大桂外科
 中田 藤重 東京都北多摩郡狛江町和泉 慈恵医大第 3 病院内科
 中田 勇 東京都杉並区高円寺 1 の 475
 中山 恒明 千葉市亥鼻町 千葉医大外科
 中山 宗春 埼玉県北足立郡戸田町新曽 2815 荘病院内
 中里 博昭 名古屋市昭和区鶴舞町 65 名古屋大学医学部今永外科
 中沢 一郎 仙台市舟丁 54
 並木 恒夫 仙台市 東北大医学病理
 南雲 秀晃 東京都港区 慈恵医大産婦人科
 南雲 昭二 東京都港区芝愛宕町 慈恵医大高木病理
 直良 博人 東京都豊島区西巢鴨 2 丁目 癌研究所内
 檜林 和之 神戸市生田区楠町丁目 神戸医科大学放射線科
 那須 健治 大阪市北区 阪大医学部第一病理
 那須 貞二 岐阜市鹿島町 岐阜市民病院
 那須 毅 松本市 信州大医学部病理
 夏目 操 岐阜市北野町 70
 根本 達久 市川市高石神 39
 二宮 一 千葉県佐原市 県立佐原病院
 西満 正 東京都文京区向ヶ丘弥生町 3 の82
 西原 武晴 札幌市 札医大内科
 西川 正夫 東京都新宿区西大久保 1 の 461 都立大久保病院
 西本 幸弘 大阪市北区 阪大医学部第一病理
 西村 光郎 東京都目黒区下目黒 4 の 863
 西村 暹 東京都豊島区西巢鴨二 癌研究所
 西村 敏雄 東京都文京区向ヶ丘弥生町 東大応微研
 新田 和男 東京都港区芝白金台町 予防衛生研

究所
西野 英雄 市川市八幡町4の1245
西牟田 祐昭 東京都大田区大森 東邦大学医学部
薬理
西尾 功 金沢市土取場永町 金沢大学医学部
外科
西尾 幸子 大阪市北区常安町 33 大阪大学医
学部第一病理
西下 秀男 岡山市 岡大医学部平木内科
西島 早見 徳島市蔵本町 徳島大学医学部第一
外科
西山 保一 川崎市新川通り70 川崎市立病院内
科
丹羽 隆 仙台市 東北大病理
野田 彰 金沢市 金沢大学医学部外科
野木 東洋 仙台市 東北大医学部桂外科
野嶽 幸雄 東京都三鷹市下連雀 41
野原 不二夫 東京都文京区本富士町 東大木本外
科
野口 貞夫 大阪市福島区堂島浜通 大阪大学医
学部久留外科
野口 義園 東京都文京区湯島天神町3の17
野内 文雄 福島市杉妻町 福島医大病理
則光 和一 熊本市城内 熊大医学部勝屋外科
野里 春夫 東京都中央区銀座東 癌研病院
野沢 勉 千葉市亥鼻町 千葉大学医学部歯科
口腔外科
額田 晋 千葉市稲毛海岸 額田医学生物学研
究所
沼田 毅 福岡市西小姓町 27 沼田病院

O

王 鍾 毓 東京都杉並区高円寺2の91
大 綱 弘 東京都文京区駒込千駄木町 日本医
大病理
小原 昭英 東京都文京区西片町 10 番地にの48
号
小原 準之助 秋田市上中城四 秋田県立中央病院
放射線科
小原 修 金沢市 金沢大第二内科
小原 辰三 東京都新宿区戸山町 国立東一病院
外科
落合 京一郎 東京都文京区 東大附属病院分院泌
尿器科
織田 昭 東京都港区芝白金台町1の39 東大
伝研

小田 博之 千葉県旭市イの1326 旭中央病院
小田 富雄 広島市千田町 広島原爆病院原爆障
碍対策研究所
小田 豊 東京都新宿区信濃町 慶応大外科
小高 健 東京都港区芝白金台町 東大伝研癌
研究所
小田島 成和 東京都千代田区神田駿河台2の2
佐々本研究所
大江 昭三 大阪市福島区 阪大泌尿器科
大藤 真 岡山市 岡山大平木内科
緒方 喜久昭 徳島市 徳島大病理
緒方 卓郎 岡山市内田 325
小川 玄一 札幌市 北大産婦人科
小川 滋 名古屋市瑞穂区 名市大第一病理
小川 勝士 岡山市岡 岡山大学医学部病理
小川 恕人 三島市谷田 1111 国立遺伝学研究所
小川 重男 東京都港区芝田村町 慈恵区産婦
人科
荻原 正雄 東京都府中市新宿 2-8192
荻野 高一 神戸市生田区 神戸医大第1内科
小倉 知治 大阪市東区法円坂町 国立大阪病院
産婦人科
大原 弘道 札幌市 札幌医大内科
大橋 伊佐治 名古屋市中区 国立名古屋病院内科
大橋 昌子 東京都豊島区西巢鴨2丁目 癌研究
所内
大橋 望彦 東京都豊島区西巢鴨2丁目 癌研究
所内
大橋 成一 東京都新宿区戸山町 国立東京第一
病院検査科病理
大平 貞雄 仙台市北四番丁 東北大学医学部黒
川内科
於保 健吉 東京都新宿区柏木 東京医大病院外
科
大星 章一 弘前市 弘前大学医学部病理
大出 浩 千葉市千葉大第二生理
及 川 淳 大阪市福島区堂島 大阪大学癌研究
所
大岩 俊夫 福岡市 九大外科
岡 治 道 東京都中野区江古田2の36
岡 博 東京都文京区本富士町1の1 東大
田坂内科
岡 隆一 大阪市北区 阪大歯学部第2口腔外
科
岡 吉彦 東京都板橋区志村前野町 81 都営ア
パート2の34 (山之内製菓)

岡田 耕一 岡山市曙町29の3
 岡田 芳彦 京都市左京区浄土寺馬場町 71 白崎嘉明方
 岡田 慶夫 京都市左京区聖護院河原町 京大結核研究所
 岡本 肇 金沢市 金沢大学医学部薬理学教室
 岡本 十二郎 東京都渋谷区原宿2の208
 岡本 達也 千葉市 千葉大病理
 岡本 敏彦 東京都文京区本富士町 東大薬学部
 岡本 吉弘 大阪市北区 阪大歯学部第2口腔外科
 岡本 良平 京都市中区西之京中保町 10
 岡村 富三 豊中市北刀根山 阪大薬学
 岡村 伸子 仙台市北四番丁 62 東北大学抗酸菌病研究所
 岡村 純 西宮市分銅町 12
 岡村 富三 豊中市北刀根山 阪大薬学部薬物学
 岡野 博光 福岡市 九大病理
 岡野 錦弥 大阪市北区 阪大病理
 岡崎 通 福岡市堅粕 九大学医学部第三内科
 岡島 邦雄 岡山市 岡大陣内外科
 大川 公康 茅ヶ崎市茅ヶ崎 10140 (国立沼津病院)
 大北 健逸 岡山市 岡山大学泌尿器科
 奥 孝一 豊中市北刀根山 382 大阪大学薬学部薬物学教室
 奥 信一 豊中市北刀根山 382 大阪大学薬学部薬物学教室
 奥田 九一郎 岡山市岡 岡山大学医学部生化学教室
 奥田 芳明 京都市左京区 京大内科
 奥 望 保 京都市左京区 京大病理
 奥村 秀夫 東京都大田区大森4の77 東邦大学医学部解剖学教室
 奥野 馨 鹿児島市 鹿児島大佐藤内科
 大森 幸夫 新潟市旭町 新潟大学医学部外科
 大向 良和 大阪市住吉区帝塚山西2の11 大阪大学微研外科
 大村 順一 岡山市 岡山大学泌尿器科
 大室 正瑞 札幌市 北大医学部産婦人科
 恩田 英司 東京都世田ヶ谷区4の751
 大根田 玄寿 東京都新宿区納戸町 13
 大西 盛光 千葉市 千葉大中山外科
 大西 俊造 大阪市北区 阪大第一病理
 小野 忠相 大阪市北区 阪大微研寄生虫部
 小野 博通 京都市左京区 京大外科
 小野 和江 東京都新宿区市ヶ谷 東京女子医大

病院産婦人科
 小野 正員 岡山市 岡大陣内外科
 小野 哲生 東京都豊島区西巣鴨2丁目 癌研究所内
 大野 隆二 熊本市本荘町 熊本大学第2外科
 大野 義雄 旭川市金星町 旭川市立病院外科
 鉄田 寛 和歌山市小松原通り 和歌山赤十字病院内科
 小野寺美津雄 千葉市 千葉大中山外科
 小野江 為則 札幌市南1条西16丁目 札幌医大病理
 小野木 昭次 東京都品川区五反田5の55 関東通信病院外科
 鬼塚 恵一郎 福岡市 九大放射線科
 織畑 秀夫 東京都新宿区河田町 10 東京女子医大外科
 折田 薫三 岡山市大供表町1の172の2
 尾崎 健次 岐阜県多治見市根木町3丁目 市営住宅ブロック3
 長村 重之 東京都新宿区柏木 東京医大内科
 長内 義章 青森県北津軽郡板柳町大町 108
 大里 俊吾 福島市 福島医科大学
 小沢 博 東京都新宿区信濃町 慶応大外科
 小沢 凱夫 大阪市住吉区 大阪府立病院
 大沢 京子 東京都新宿区大京町 27
 合瀬 義晴 佐賀市大財町 209 の3
 尾関 己一郎 久留米市 久留米大放射線科
 尾島 昭次 京都市左京区 京都大学医学部病理
 大島 秀彦 津市三重大公衆衛生学
 大島 桂太 名古屋市昭和区御器所町1の10
 大島 康夫 東京都墨田区平川橋5の7 第一製薬研究所
 大下 寿隆 東京都大田区大森 東邦大学第一内科
 大 蘭 卓 東京都板橋区志村小豆沢町 山之内製薬内
 太田 武八郎 大阪市東区道修町 武田薬品工業開発課
 太田 忠造 弘前市 弘前大頼外科
 太田 和雄 名古屋市 名大日比野内科
 太田 邦夫 東京都文京区指ヶ谷町 90
 太田 善介 岡山市 岡山大学平木内科
 大竹 久 岩手県盛岡市 岩手医大放射線医学
 大谷 博 京都市左京区修学院仏者町 12
 尾辻 省吾 鹿児島市 鹿児島大佐藤内科
 大塚 久 福岡市 九大医学部病理

大槻 菊男 東京都文京区小石川丸山町 19
大内 謙二 仙台市 東北大病院
大山 治史 鹿児島市山下町

P

Prince, Alfred M.

Department of Virus and Rickettsial Diseases
Medical General Laboratory (406)
Kamitsuruma (上鶴間)
Sagamihara-shi (相模原市)
Kanagawa-ken.

R

李 英 徹 韓国 什魯 特別市鎧路区鎧路三街 62
六城 雅 彙 東京都豊島区千川町 1 の 16
六尾 勝美 名古屋市昭和区 名大今永外科

S

西条 力男 仙台市北四番丁 東北大学医学部黒川内科
斎藤 寿 熊本市本荘町 熊大第 2 外科
斎藤 博 山形市半郷国保直営蔵王半郷診療所
斎藤 宏 新潟市旭町 新潟大学医学部外科
斎藤 守 東京都豊島区西巢鴨 2 丁目 癌研究所
斎藤 満 東京都北区田端 805 東鉄衛生試験室
斎藤 昌介 東京都大田区 東邦大薬理
斎藤 達雄 仙台市堤通 35
斎藤 武郎 福島市杉妻町 福島医大病理
斎藤 敏明 東京都新宿区信濃町 慶応病院
三枝 達明 大阪市北区 阪大微研病院
佐伯 政雄 東京都北多摩郡保谷町 639
相模 成一郎 大阪市福島区 阪大病院皮膚科
坂土 一英 福岡市 九大第 2 外科
酒井 英二 東京都港区芝愛宕町 慈恵医大産婦人科
酒井 克治 大阪市阿倍野区旭町 大阪市立大学部自羽外科
酒井 清周 塩釜市立病院
堺 哲郎 新潟市旭町 新潟大学医学部外科
榊 欽也 枚方市大字坂 275 大阪歯大生化学
榊原 栄一 大阪市天王寺区南河堀町 大阪学芸大学微生物学研究所

榊原 道雄 東京都中央区銀座東 癌研病院
榊原 宜 岡山市北方中井町 722
坂元 純郎 東京都渋谷区幡ヶ谷原町 780
坂本 武司 岡山市西中山下 89 財団法人川崎病院川崎癌研究所
坂本 恒雄 東京都文京区西片町 10 のいの 26
佐川 文明 東京都品川区平塚 6 の 1016 昭和医大第二病院
坂上 良男 東京都文京区向ヶ丘弥生町 東大応微研
崎田 重康 金沢市 金沢大学医学部第 2 内科
佐久間 貞行 名古屋市昭和区 名大放射線医学
桜木 四郎 東京都新宿区戸山町 国立第一病院放射線
桜井 孝 宇部市東区明神町 3 丁目
桜井 欽夫 東京都北区西ヶ原 1 の 26 薬理研究所
桜庭 司 青森市八重田字浜野 22 の 1
桜根 好之助 大阪市西区江戸堀上通り 2 の 42 金原商店方
鮫島 博 福岡県甘木市 県立朝倉病院皮膚科
鮫島 啓二郎 東京都文京区 東大医学部分析
鮫島 夏樹 札幌市北 14 条西 5 丁目 北大病院第 2 外科
鮫島 哲也 鹿児島市 鹿児島大佐藤内科
三戸 康郎 福岡市堅粕 九大第 2 外科
山東 正昭 東京都新宿区信濃町 慶大外科
佐々 弘 東京都新宿区東大久保 東京医大病院
笹井 憲三 豊岡市立野 公立豊岡病院
笹井 外喜雄 京都市右京区嵐山中尾下町 54
佐々木 本道 札幌市北 10 条西八丁目 北大理学部動物学教室
佐々木 常雄 名古屋市昭和区 名大放射線医学
佐々木 義徳 仙台市北四番丁 東北大学医学部内科
笹野 伸昭 仙台市 東北大病理
佐野 開三 岡山市岡 岡山大学病院砂田外科
佐野 量造 青森市 県立中央病院病理検査科
指田 勢郎 東京都新宿区柏木 東京医大外科
佐曾利 孝 名古屋市中区新栄町 名古屋大学附属病院分院外科
佐竹 実 札幌市 札幌大産婦人科
佐藤 秩子 名古屋市昭和区名大病理
佐藤 八郎 鹿児島市 鹿児島大内科
佐藤 春郎 仙台市 抗酸菌病研究所

- 佐藤 寿昌 名古屋瑞穂区 名市大病理
佐藤 博 東京都世田谷区粕谷町 39
佐藤 博 東京都北区西ヶ原1の26 薬理研究
所内
佐藤 宏 仙台市 東北大学病院桂外科
佐藤 裕信 東京都品川区西品川1の 888 三共
株式会社研究部
佐藤 伊吉 千葉市玄鼻町 312 千葉大学医学部
歯科口腔外科
佐藤 仁 弘前市相良 弘前大医学部第一外科
佐藤 勝己 札幌市南1条西 16 丁目 札幌医大
病内科学教室
佐藤 和男 仙台市北四番丁 62 東北大学抗研
佐藤 健二 東京都世田ヶ谷区深沢町1の 899
東京都立アソートブ総合研究所第
4部
佐藤 研介 札幌市 札幌医大内科
佐藤 清 東京都杉並区柿木町 144
佐藤 正弘 仙台市 東北大抗研
佐藤 光永 弘前市 弘前大学病理
佐藤 宮彦 仙台市 東北大桂外科
佐藤 端 岡山市上伊福 71
佐藤 永雄 東京都品川区平塚 昭和医大生化学
教室
佐藤 七郎 鎌倉市大町 778
佐藤 拓司 札幌市 北大第2外科
佐藤 徳郎 東京都杉並区阿佐ヶ谷1の 861
佐藤 東正 千葉市 千葉大学産婦人科
佐藤 登志郎 東京都文京区本富士町 東大物療内
科
佐藤 隆一 群馬県邑楽郡大泉町大字上小泉2310
佐藤 麟太郎 岩手県江刺市 県立江刺病院
佐藤 泰雄 岡山市岡 164 岡山大学病院砂田外
科
佐藤 順泰 東京都中央区銀座東 癌研病院
沢田 平十郎 大阪市阿倍野区 阪市大沢田外科
沢田 秀作 大阪市住吉区万代東2の33
沢山 恵美子 三重県志摩郡大王町波切 県立大王
病院産婦人科
清 英夫 西宮市相生町 157
西海 雄幸 福岡市 九大病理
清家 澄保 京都市左京区 京大外科
瀬木 三雄 仙台市 東北大衛生学
関 一子 東京都大田区 東邦大薬理
関 健次郎 鎌倉市小町 287
関 正利 千葉市玄鼻町 千葉大学医学部病理
関口 守正 東京都文京区駒込西片町 10 のへの
17
関口 豊三 千葉市黒砂町放医研障害基礎研究部
関矢 偲 新潟市旭町 新潟大学医学部外科
関山 重孝 東京都港区芝愛宕町 慈恵医大高木
病理
仙石 光彦 名古屋市昭和区鶴舞町 名古屋大学
医学部今永外科
瀬辺 恵鎧 熊本市城内 熊本大医学部第二薬理
妹尾 左知丸 岡山市国富 16
瀬戸 淳一 仙台市 東北大黒川内科
瀬戸 秀一 仙台市光禅寺通 35
芝 茂 大阪市北区堂島西町 7 大阪大学微
生物研究所外科
柴田 凡夫 岡山市岡 164 岡山大学医学部平木
内科
志田 正夫 佐賀市水ヶ江町桜小路 16
重野 荔 札幌市 北大産婦人科
重富 正三 宇部市西区松島町二丁目
椎名 栄一 千葉市花園町 75
志方 俊夫 東京都文京区 東大病理
島 隆允 米子市西町 鳥取大学医学部放射線
医学教室
島田 洋 熊本市 熊本大産婦人科
島田 甚晴 大阪市浪速区逢坂下之町 20 島田
医院
島田 馨 東京都文京区 東大田坂内科
島田 信勝 東京都杉並区阿佐ヶ谷3の 504
島本 雄一 神戸市生田区 神戸医大放射線科
島村 嘉高 東京都江戸川区平井1の 1631
嶋崎 昌義 和歌山市 和歌山医大病理
嶋津 孝 大阪市福島区 阪大癌研
清水 浩 東京都文京区 日本医大放射線医学
清水 準也 岡山市 岡大陣内外科癌研
清水 進 川崎市片平 195
清水 保 東京都文京区本富士町 東京大学医
学部物療内科
清水 康世 岐阜市司町 岐阜医大乾内科
下山 利雄 札幌市 札幌大産婦人科
品川 信良 弘前市本町 53 弘前大学医学部附
属病院産婦人科
新保 幸太郎 札幌市南一条西 17 丁目 札幌医大
病理
新宮 雅 大阪府泉南郡南海町尾崎 102 上田
方
篠原 護 札幌市 札幌大産婦人科

篠井 金吾 東京都新宿区柏木 東京医大外科
 篠木 隆男 福島市杉妻町 福島医大遠藤外科
 篠崎 忠吉 東京都江戸川区小岩町4の1950
 篠崎 一雄 東京都港区芝愛宕町 慈恵医大産婦人科
 篠崎 達世 弘前市 弘前大放射線科
 篠沢 貞夫 東京都荒川区日暮里町2の218
 汐見 文隆 和歌山市小松原通り 和歌山赤十字病院第1内科
 塩谷 卓爾 東京都神田駿河台2の2 杏雲堂病院
 白羽弥右衛門 大阪市阿倍野区 阪市大白羽外科
 白石 彰徳 岡山市岡 岡山大学医学部平木内科教室第二研究所
 白須 泰彦 東京都豊島区西巢鴨2丁目 癌研究所
 白沢 健二郎 松本市 信州大病理
 蘇 珩山 中華民国台湾省高雄市 塩埕区健国四路 135
 副島 一彦 福岡市堅粕 九大第2外科
 添田 博彬 福岡市堅粕 九州大学医学部放射線医学
 曾我 淳 新潟市自由ヶ丘 1240 の51
 相馬 智 熊谷市大字熊谷 904
 相馬 広明 東京都台東区向柳原町2〜1 大西方
 染谷 守 浦和市針ヶ谷1の53
 園田 孝夫 大阪市東住吉区山坂町3の43
 外村 昭 札幌市 北大理学部動物
 外野 正己 東京都新宿区東大久保 東京医大病理
 須田 正己 大阪市西区土佐堀前通り3の22
 大阪大学蛋白質研究所
 末松 俊彦 大阪市東淀川区野中南通1の63
 須賀 昭二 愛知県江南市庵子島
 菅 邦彦 京都市上京区河原広小路 京都府立医大病理
 菅原 道義 弘前市 弘前大病理
 菅井 孝 東京都板橋区大谷口上町 日大細菌学
 菅野 晴夫 東京都文京区 東大医学部病理
 菅野 孝一 福島県伊達郡月館町大字月館字町30
 菅野 巖 仙台市 東北大抗研
 杉 義吉 福岡市堅粕 九州大学医学部第一内科
 杉原 克己 仙台市 東北大病理

杉原理 一 奈良県大和高田市大字高田 1573
 杉本 顕俊 大阪市北区常安町 大阪大学医学部第二病理
 杉本 年 四日市市塩浜町 三重県立大学医学部附属塩浜病院
 杉本 毅 東京都文京区 東大産婦人科
 杉村 隆 東京都豊島区西巢鴨2丁目 癌研究所
 杉谷 幸男 東京都品川区西品川1の888 三共株式会社研究部
 杉山 浩太郎 福岡市鳥飼町6の499
 杉山 正 東京都新宿区戸山町 東一病院癌相談室
 砂田 輝武 岡山市 岡山大砂田外科
 角 南 宏 岡山市 岡山大学平木内科
 鈴江 懐 京都市左京区 京大病理
 鈴木 千賀志 仙台市 東北大学抗酸菌病研究所
 鈴木 博孝 千葉市 千葉大中山外科
 鈴木 鍾美 東京都千代田区三崎町 東京歯科大病理
 鈴木 恵之助 千葉市亥鼻町 千葉大学医学部中山外科
 鈴木 実 仙台市 東北大病理
 鈴木 三郎 東京都港区赤坂葵町2 東京電気通信局内
 鈴木 慎二 横浜市港北区箕輪町 501
 鈴木 修一 東京都中野区住吉町 25 富士見荘 10号
 鈴木 忠雄 東京都中央区銀座東 癌研病院
 鈴木 竜哉 愛知県蒲郡市蒲郡町中沢9
 鈴木 善久 仙台市 東北大黒川内科

T

田端 敏秀 大阪府貝塚市近木町 978
 田淵 昭 広島市広大産婦人科
 田淵 幸博 大阪市北区堂島西町3 大阪大学微生物研究所病院
 橋 貞亮 金沢市 金沢大第1外科
 橋 芳郎 仙台市 東北大病理
 立花 省吾 岡山市岡 岡山大学医学部産婦人科
 立入 弘 大阪市福島区 阪大病院放射線科
 立沢 定直 東京都葛飾区表戸町1の998 葛飾結核検診病院
 多田 秀夫 枚岡市額田町 1956
 多田 正俊 高松市五番丁
 田頭 勇作 京都市左京区吉田近衛町 京大ウイ

ルス研究所病理		高岡 満	東京都世田谷区玉川奥沢町1の44
多ヶ谷 勇	東京都品川区上大崎長者丸 国立予研リケッチャウイルス部	高岡 聴子	東京都港区芝白金台町 伝染病研究所病理
多胡 健吾	岡山市岡 岡山大学医学部津田外科	高瀬 貞夫	仙台市 東北大病理
田郷 寿正	東京都中野区本郷通1の42	高島 啓昌	東京都大田区 東邦大薬理
田口 鉄男	大阪市北区堂島西町3 大阪大学微生物病研究所外科	高島 正敏	東京都港区芝愛宕町 慈恵大産婦人科
高木 文一	東京都港区芝愛宕町 東京慈恵会医大病理	多賀須 幸男	東京都文京区本富士町 東京大学医学部田坂内科
高木 忠一郎	長崎市坂本町 長崎医大産婦人科	高杉 年雄	札幌市 北大第三内科
高木 弘	東京都品川区西品川1の888 三共高峰研究所	高柳 伊立	倉敷市美和町 倉敷中央病院研究所
高木 国夫	東京都中央区銀座東 癌研病院	高山 欽哉	神奈川県足柄郡山北町 舟水医院
高木 良三郎	福岡市堅粕 九州大学医学部第一内科	高山 昭三	東京都豊島区西巢鴨2 癌研究所内
高橋 文子	東京都新宿区市ヶ谷河田町 東京女子医大病院産婦人科	高山 担三	札幌市南1条西17丁目 札幌医大外科
高橋 等	熊本市本荘町 熊本皮膚科	高山 奨	西宮市上ヶ原 関西学院大学理学院生物学
高橋 淳	東京都世田谷区弦巻町1の7	武田 勝男	札幌市 北大病理
高橋 健一	東京都練馬区東大泉町 834	武田 進	津市 三重大病理
高橋 皓	函館市会所町 55	武田 敏	東京都江戸川区小松川3の38
高橋 希一	仙台市 東北大外科	竹田 斌郎	奈良市東本辻町7
高橋 理明	大阪市北区堂島西町 大阪大学微生物研究所ウイルス学部	武田 俊輝	岡山市岡 岡山大学医学部平木内科
高橋 学	宇部市 山口医大病理	武田 義章	大阪市 北区阪大外科
高橋 勝	京都市左京区 京大医学部外科	竹原 靖明	東京都新宿区戸山町 国立東一病院外科
高橋 良吉	前橋市岩神町 群馬放射線科	武石 輝夫	東京都大田区田園調布3~134
高橋 信次	名古屋市中昭和区名大放射線	武正 勇造	東京都杉並区高円寺1の27
高橋 哲也	福岡市 九大第一内科	竹本 和夫	東京都中央区日本橋両国 34
高橋 泰常	福岡市堅粕 九州大学医学部癌研究生化学	竹村 敏朗	東京都新宿区上落台1の466 やよい荘8号
高橋 富	仙台市通町 282	竹島 和夫	熊本市 熊本大産婦人科
高井 新一郎	大阪市生野区勝山通9の72	竹島 竹	福島市杉妻町 福島医大病理
高井 新治	東京都世田谷区池尻町 自衛隊中央病院研究部	竹内 純	名古屋市昭和区鶴舞町 名古屋大学医学部第二病理
高川 幹男	仙台市 東北大黒川内科	竹内 清	名古屋市昭和区鶴舞町 名古屋大学医学部第一内科
高松 英雄	京都市左京区 京大結核研究病理	竹内 正七	東京都文京区本富士町1 東京大学医学部産婦人科教室
高松 勇雄	福岡市 九大病理	竹内 正	東京都板橋区大谷口町 日大医学部病理
高宮 和彦	東京都文京区東大農芸化学 住木研究室	武内 忠男	熊本市本庄町 熊本大病理
高野 宏一	東京都荒川区尾久町5の1135 佐藤病院内	竹内 富雄	東京都港区芝白金台町 国立予防衛生研究所
高村 正衛	東京都品川区五反田5の55 関東通信病院	滝 一郎	大阪市南区千年町 14
高村民雄	札幌市南1条西16丁目 札幌医大内科学教室	滝川 清治	名古屋市中区梅枝町2の7
		田北 周平	徳島市蔵本町 徳島大第1外科

- 田北 暉比呂 福岡市堅粕 九大医学部放射線医学
滝本 庄蔵 札幌市南1条西14丁目 札幌医大病院内科
滝野 義忠 大阪市東住吉区平野西脇町18 大日本臓器研究所
滝沢 延次郎 東京都文京区駒込曙町12
玉井 恭子 福岡市堅粕 九州大学医学部病理
玉井 定美 弘前市在府町5 弘前大医学部病理
玉木 正男 長崎市坂本町 長崎大放射線科
玉真 俊一 千葉市 千葉大中山外科
玉利 彰 東京都港区芝愛宕町 慈恵大産婦人科
田村 潤 名古屋市中村区広路町 松風園9
田村 晃 東京都台東区浅草寿町3の24
田村 宏平 東京都世田谷区玉川上野毛町392
田村 竜男 東京都中央区銀座東 癌研病院
田辺 秀治 弘前市 弘前大医学部病理
田中 宏 大阪市東成区森町南1丁目 大阪府立成人病センター
田中 勝治 東京都新宿区信濃町 慶応大外科
田中 啓真 鎌倉市乱橋材木座1233
田中 健之介 川崎市大島町4の52
田中 晃 東京都港区 慈恵医大産婦人科
田中 邦喜 大阪市東淀川区十三西之町4 武田研究所内
田中 昇 東京都渋谷区宮代町 日赤中央病院病理
田中 信夫 東京都渋谷区代々木初台512 武田方
田中 信男 東京都文京区向ヶ丘弥生町 東大応微研
田中 信徳 東京都文京区本富士町 東京大学理学部植物学教室
田中 正明 奈良市中筋町13
田中 治 和歌山市七番町 和歌山医大第一内科
田中 早苗 岡山市 岡山大学陣内外科
田中 昭平 東京都台東区浅草柳橋2の4
田中 聡 岡山市岡154 岡山大学医学部津田外科
田中 聖児 宇部市車見初 車見初病院外科
田中 伸一 名古屋市中区長堀町3の10
田中 達也 札幌市北八条 北大理学部動物学教室
田中 富子 東京都豊島区西巣鴨2 癌研究所内
田中 俊雄 岡山市上伊福中町661の6 草地方
高橋 嗣明 弘前市本町1の33 安井栄蔵方
田中 義良 名古屋市中区南外堀町 国立名古屋病院内科
種子田 哲郎 鹿児島市山下町 鹿児島大第2内科
谷川 精一 福岡市 九大外科
谷戸 健蔵 旭川市 旭川赤十字病院内科
立岩 道正 名古屋市中区新栄町3の29 名古屋大学医学部分院外科
立岡 末雄 大阪市東淀川区十三西之町 武田研究所
田坂 定孝 東京都文京区本富士町 東大田坂内科
田代 勝洲 名古屋市中村区元中村町1の30
田坂 純雄 岡山市 岡山大泌尿器科
田代 仁男 熊本市城内 熊本大医学部産婦人科
田代 田鶴子 東京都北区西ヶ原1の26 薬理研究会研究所
建部 守昭 金沢市土取場永町 金沢大学医学部第二病理
田内 久 名古屋市中区昭和区鶴舞町 名古屋大学医学部病理
田崎 英生 東京都渋谷区豊沢町68 都立広尾病院放射線
田崎 勇三 東京都大田区上池上町51
田沢 多郎 東京都港区 慈恵医大産婦人科
手島 貞一 仙台市 東北大病理
天神 美夫 東京都中央区銀座東 癌研病院
寺畑 喜朔 金沢市下石引町 国立金沢病院外科
寺尾 清 千葉市 千葉大病理
寺島 寛 大阪市阿倍野区旭町 大阪市立大学医学部病理
寺島 芳輝 東京都港区芝愛宕町 慈恵医大産婦人科
寺下 樽治 大阪市北区堂島西町3 大阪大学微生物病研究所病院
寺沢 敏夫 大阪市福島区堂島浜通 大阪大学医学部附属病院第二外科
寺山 宏 東京都文京区本富士町 東京大学理学部化学
勅使河原 巍 東京都中央区日本橋本町4の6 興和化学研究所
寺脇 朝治 大阪市北区堂島西町 大阪大学微生物研究所外科
戸木田 菊地 東京都大田区 東邦大薬理
徳川 博武 東京都目黒区上目黒8の521
徳永 昭夫 大阪市福島区堂島浜通 大阪大学医学部久留外科

- 徳岡 淳一 東京都千代田区神田駿河台 2 の 2
杏雲堂医院
- 徳岡 俊次 京都市左京区下鴨北園町 108 の 1
- 徳山 英太郎 東京都新宿区若松町 41
- 友尻 諒弥 熊本市本莊町 熊大第二外科
- 富岡 宏行 岡山市 岡山大砂田外科
- 泊 康男 金沢市金沢第二内科
- 都丸 禎三 東京都豊島区西巢鴨 2 癌研究所内
- 渡守武 健 大阪市福島区海老江上 2 の 1 大日本製薬中央研究所
- 戸田 智博 福岡市 九大外科
- 虎谷 良雄 和歌山市七番町 和歌山医大第一内科
- 登坂 邦雄 東京都大田区 東邦大薬理
- 豊島 克 東京都文京区 東大産婦人科
- 戸嶋 シマエ 東京都世田谷区太子堂太子堂住宅52
- 十束 支朗 市川市新田町 1 の1136 菅各方
- 豊田 滋夫 徳島市蔵本町 徳島大第一外科
- 津田 福視 福島市杉妻町 福島医大第一内科
- 津田 文男 茨城県猿島郡総和村 猿島日赤病院
- 津田 一彦 東京都台東区御徒町 2 の65
- 津田 豊彦 西宮市清水町 12
- 辻 由生子 札幌市北 12 条西 5 丁目 北大医学部第一病理
- 辻井 平三 尼崎市今福 192 塩野義研究所
- 辻本 宏 橿原市畹傍 奈良医大病理
- 塚田 英之 札幌市南 1 条西 17 丁目 札幌医大病理
- 塚越 茂 東京都練馬区豊玉北 4 の26
- 塚本 憲甫 市川市菅野 4 の 1226
- 塚崎 鴻 東京都新宿区柏木 東京医大外科
- 常松 匠 東京都千代田区神田駿河台 2 の 2
杏雲堂病院外科
- 坪井 栄孝 東京都文京区駒込動坂町 339 日本医大附属医院放射線科
- 坪井 香容子 名古屋市南区戸部町 3~52
- 螺良 義彦 橿原市 奈良医大病理
- 土田 亮一 富山市西長江 県立中央病院内科
- 綱村 史郎 金沢市 金沢大第一外科
- 恒元 博 金沢市下石引町 国立金沢病院放射線科
- 津島 恵輔 弘前市 弘前大桂外科
- 堤 啓 岡山市 岡山大学病理
- 都築 正志 名古屋市北区楠町大字味鏡字茨道 5169 の 1
- 都築 俊治 埼玉県越谷市越ヶ谷 702
- 筑紫 清太郎 東京都港区赤坂葵町 虎ノ門病院外科
- 都築 敏男 大阪市北区 阪大微研病院
- U
- 内田 一 金沢市九人橋下通 3 内田病院
- 内田 耕太郎 京都市左京区 京大医学部外科
- 内田 正男 東京都世田谷区玉川瀬田町 836
- 内田 洋子 東京都新宿区市ヶ谷 東京女子医大病院産婦人科
- 内田 豊 岡山市西中山下 財団法人川崎病院川崎癌研究所
- 内海 耕造 岡山市 岡山大癌研
- 内野 晃 東京都新宿区信濃町 慶応大外科
- 内野 治人 京都市左京区松ヶ崎西桜木町 10
- 内山 節夫 大阪市福島区堂島 大阪大学医学部癌研究所
- 宇田川 啓次 仙台市 東北大抗研
- 宇田川 康也 東京都新宿区信濃町 慶応大外科
- 上田 憲次 室蘭市東町 40 の 7
- 植田 健治 佐賀市水ヶ江町 佐賀県立病院好生館産婦人科
- 上田 幸藏 長野県伊那松島 9808 西沢生化学研究所
- 上田 正規 大阪市 北区阪大微研外科
- 植木 寛 熊本市大江町 熊大薬学部第一製薬
- 上村 光夫 鹿児島市山下町 70 鹿児島大学附属病院第二内科
- 上村 良一 広島市出沙町 681
- 上西 力 大阪市北区 阪大微研外科
- 上野 明 東京都文京区 東大木本外科
- 上埜 忠一 北海道函志郡熊石村 熊石第二診療所
- 鶴飼 光雄 名古屋市昭和区 名大今永外科
- 鶴飼 典司 小牧市小牧 1823 小牧病院
- 浮島 仁也 東京都文京区本富士町 東大医学部木本外科
- 馬屋原 晟 福岡市 九大放射線医学
- 梅田 誠 東京都港区芝白金台町 東大伝研
- 梅田 真男 東京都豊島区池袋東 1 の81 一ふじ荘
- 梅原 裕 弘前市 弘前大病理
- 梅村 慎一郎 東京都板橋区大谷口町 日本大学医学部病理
- 梅沢 浜夫 東京都港区芝白金台町 国立予研
- 梅園 忠 東京都武蔵野市吉祥寺町 600

宇野 広治 弘前市 弘前大学医学部第一病理
 宇野 裕 名古屋市昭和区鶴舞町 名古屋大学
 医学部病理
 魚住 光洋 大阪市福島区堂島 大阪大学医学部
 癌研究所
 卜部 美代志 金沢市 金沢大学医学部卜部外科
 浦田 吟夫 東京都板橋区 日大板橋病院第一外
 科
 浦口 健二 東京都目黒区中目黒4の1の221
 白 淵 勇 弘前市 弘前大学医学部病理
 白井 茂樹 大阪市福島区 阪大泌尿器科
 白居 敏仁 大阪市東淀川区十三西之町 武田研
 究所
 牛 島 宥 名古屋市昭和区 名大中央臨研病理
 牛尾 暉夫 東京都足立区五兵衛町 77
 後 昭 行 福岡市堅粕 九大外科
 牛山 篤夫 長野県茅野市宮川 市立茅野町病院
 宇津木 和夫 東京都世田谷区世田谷 3の2362
 宇都宮 英一 福岡県田川郡金田町
 宇都宮 讓二 東京都世田谷区玉川尾山町 130

W

和田 昭 大阪市北区常安町 大阪大学医学部
 第一病理
 和田 薫 札幌市 札幌医大内科
 和田 典之 堺市南三国丘町 1の26
 和田 武雄 札幌市南1条西 16丁目 札幌医大
 病院内科
 和田 卓人 福岡市 九大病理
 和田 義夫 名古屋市中区 国立名古屋病院内科
 若林 克己 東京都文京区 東大薬学生理化学
 若林 勝 札幌市北 14条5丁目 北大医学部
 放射線医学
 若林 修 東京都板橋区大谷町 日大医学部
 外科
 若狭 一夫 宮城県柴田郡船岡 鉄道病院
 若狭 毅 仙台市 東北大病理
 若佐 理 新潟市旭町 新潟大学医学部外科
 若杉 広太郎 西宮市染殿町 60 西宮市民病院
 若木 重敏 東京都千代田区有楽町 1の6 協和
 醸酵工業株式会社技術部研究課
 脇坂 行一 京都市左京区吉田 京都大学医学部
 第一内科
 鷺見 敏 名古屋市熱田区外土居町 48 新三
 菱名古屋病院産婦人科
 渡辺 文友 長崎県大村市徳泉河内郡 長崎大学

医学部家畜医学研究所
 渡辺 弘 東京都千代田区神田和泉町 三井厚
 生病院外科
 渡辺 巖 大阪市 阪大第一外科
 渡辺 憲市 金沢市 金沢大第一外科
 渡辺 三喜男 神戸市灘区高羽鷹匠 6の5
 渡辺 命平 熊本市 熊本大産婦人科
 渡辺 貞雄 宇部市宇部山口医大
 渡辺 昭一 福島県相馬郡鹿島町横手字川原 2
 鹿島厚生病院
 渡辺 修治 箕面市箕面 431
 渡辺 武夫 浜田市殿町 浜田通信診療所
 渡辺 民朗 仙台市 東北大抗研
 渡辺 俊夫 山形市香澄町桜小路 篠田病院第二
 外科
 渡部 恒 仙台市 東北大武藤外科
 渡辺 順明 大阪市東淀川区加島町 371 武田薬
 工社宅
 渡辺 良子 大阪市生野区林寺町 2~65
 渡辺 善正 大阪市北区 阪大微研病院
 綿貫 勤 仙台市 東北大病理
 亘理 健一 仙台市 東北大黒川内科
 巖岡 小太郎 東京都豊島区西巢鴨 2 癌研究所内

Y

矢川 寛一 盛岡市内丸 岩手医大病理
 八木 日出雄 岡山市 岡山産婦人科
 八木 力 仙台市北四番丁 東北大学医学部
 黒川内科
 八木 泰夫 金沢市 金沢大第二内科
 山田 裕 札幌市 札幌医大産婦人科
 山田 文夫 大阪市阿倍野区 阪市大産婦人科
 山田 一正 名古屋市昭和区 名大日比野内科
 山田 耕司 東京都文京区 東大産婦人科
 吉田 弘一 仙台市元寺小路 92
 山田 喬 東京都千代田区神田駿河台 2の2
 佐々木研究所
 山田 真喜雄 貝塚市橋矢 1587 国立大阪療養所
 山田 正篤 東京都中央区日本橋茅場町 2の3
 山田 光雄 東京都大田区 東邦大薬理学
 山田 肅 東京都中央区銀座東 癌研病院
 山田 敏郎 東京都文京区 東大理学部生化学寺
 山研究室
 山田 欽 東京都杉並区下高井戸 1の108
 山形 敏一 仙台市 東北大山形内科
 山県 英士 東京都新宿区戸山町 35 ときわ荘1

- 山口 淳二 仙台市 東北大抗研
山口 巖 仙台市 東北大武藤外科
山口 寿 京都市北区平野島居前 37
山口 健二 尼ヶ崎市今福 塩野義製薬研究所
山口 金吾 仙台市 東北大武藤外科
山口 延男 京都市左京区 京大腸坂内科
山口 清三 大阪市北区 阪大微研病院
山口 康夫 東京都板橋区大谷口町 日本大学医学部細菌学教室
山川 真 大阪市福島区 阪大病院武田外科
山川 拓 福岡市 九大第二外科
山岸 一一 川崎市小杉町1の396 日本医大第二医院
山本 裕彦 新潟市 新潟大医学部病理
山本 研二郎 尼ヶ崎市今福 192 塩野義製薬抗瀬工場研究所
山本 清 和歌山市小松原通 和歌山赤十字病院第一内科
山本 祐夫 大阪市阿倍野区旭町1の61 大阪市立大学医学部石井内科
山本 道夫 岡山市 岡山大癌研
山元 清一 名古屋市東区東芳野町1の90
山本 正 東京都港区芝白金台町1の39 東京大学伝染病研究所第五細菌研究室
山村 雄一 福岡市 九州大学医学部医化学
山名 寛治 米子市 鳥取大桑原外科
山中 晃 東京都豊島区椎名町2の1852
山中 豊 西宮市大畑町 22 前原方
山宮 克己 新潟市 新潟大医学部
山岡 憲二 福岡市堅粕 九州大医学部第一内科
山岡 静三郎 大阪市東区北浜5 住友化学工業技術部
山崎 光郎 東京都大田区田園調布4の47の18
山崎 可夫 市川市八幡町4の1238
山下 久雄 東京都豊島区西巢鴨2 癌研究所内
山下 貢司 宇部市 山口医大病理
家森 武夫 神戸市生田区楠町 神戸医大第一病院
安江 正子 岡山市 岡山県庁内 岡山衛生部公衆衛生科
安河内 秀幸 神戸市生田区神戸医大病理
安村 美博 千葉市 千葉大細菌学
安岡 正輝 東京都世田谷区池尻自衛隊衛生学校
柳田 隆穂 大阪市阿部野区阪南町1の57
柳本 行雄 大阪市東区京橋1丁目 大阪歯科大
柳沢 文憲 千葉市 千葉大中山外科
矢野 久雄 大阪市福島区 阪大病院 泌尿器科
矢野 潔 鳥取市 鳥取赤十字病院放射線科
余 語 弘 名古屋市昭和区陶生町1の10
横路 謙次郎 広島市 広大病理
横山 秀吉 名古屋市中区7本松町2の13
横山 育三 京都市左京区 京大医学部外科
横山 哲夫 千葉市 千葉大中山外科
米田 光作 大阪市福島区 阪大第一外科
米田 友彦 大阪市東淀川区十三西之町 武田研究所
葉 緑 孫 東京都新宿区戸塚町4丁目 戸山アパート 10の225
吉田 耕 東京都大田区小林町 296
吉田 穰 名古屋市昭和区鶴舞町 名古屋大学医学部今永外科
吉田 順之助 弘前市 弘前大病理
吉田 武彦 東京都千代田区富士見町2の3 東京警察病院産婦人科
吉田 竹瑯 東京都豊島区堀之内 984
吉岡 鉄郎 島根県大原郡大東町 雲南共存病院 官舎
吉田 富三 東京都文京区本富士町 東京大学医学部病理学教室
吉田 俊秀 三島市 国立遺伝学研究所
吉田 良行 京都市左京区岡崎北御所町 42の11
吉浜 博太 東京都品川区五反田5の55 関東通信病院
吉川 春雄 北海道空知郡中富良野村国保病院内
吉村 克俊 東京都品川区五反田5の55 関東通信病院
吉村 輝久雄 大阪市泉北郡信太村字太 171
吉村 義之 横浜市南区 横市大病理
善 成 務 大阪市福島区堂島浜通り 大阪大学医学部放射線科
吉永 直胤 熊本市本荘町 熊本大二外科
吉岡 俊一 橿原市畝傍 奈良医大病理
吉岡 濟 大阪市北区 阪大歯学第一口腔外科
吉武 泰男 東京都品川区五反田5の55 関東通信病院
湯浅 録介 東京都大田区桃谷町2の730
湯川 永洋 大阪市東区今橋3 湯川胃腸病院
柚木 一雄 鹿児島市山下町 70 鹿児島大学医学部内科

日本癌学会会則

(Bylaw of the Japanese Cancer Association)

第 I 章 名 称

第 1 条 本会は、日本癌学会と称する。

第 II 章 目的及び事業

第 2 条 本会は、癌研究の発達を図ることを以つて目的とする。

第 3 条 本会は、前条の目的を達成するために次の事業を行う。

1. 学会総会及び其他の学術的集会
2. 機関誌の発行
3. その他

第 4 条 本会事務所は、東京都豊島区西巣鴨 2 丁目 2615 番地癌研究所内に置く。

第 III 章 会 員

第 5 条 本会評議員 1 名の紹介状を附して、本会事務所に入会を申込み会費を納入するものは会員となり得る。

第 6 条 会員は、毎年 3 月末日までに事務所に会費を前納する義務がある。

第 7 条 会員は、機関誌の配布をうけ、且つ本会学会総会に出席して、業績を発表し発言することが出来る。

第 8 条 会員が退会又は転居する場合は、事務所に通知しなければならない。二年以上引つづき会費を滞納し督促に応じない場合その他本会員としての名誉を傷つけた場合は評議員会の決議によつて退会せしめることができる。

第 9 条 本邦癌学界のために多大の貢献をなしたものは、評議員会の決議に基いて名誉会員に推薦されることがある。

第 IV 章 役 員

第 10 条 本会に会長、副会長各 1 名、幹事若干名及び評議員 75 名以内の役員を置く。

第 11 条 会長は、幹事の互選によつて定められ、本会を主宰しこれを代表する。会長の任期は一年とし再選を妨げない。

第 12 条 副会長は幹事の中から会長これを委嘱する。副会長は、会長を補佐し且つ其の代理を行う。副会長の任期は一年とし再選を妨げない。

第 13 条 幹事は評議員の互選によつて定められ、会務に関する重要事項を協議し実行する。幹事の中から庶務、会計、及び編集を掌るもの各 1 名を互選する。幹事の任期は二年とし再選を妨げない。

第 14 条 評議員は、継続三十年以上の会員であつて研究機関の公職にあるものの中から幹事会の推薦に基いて毎年会長が委嘱する。評議員は評議員会を組織し本会に関する重要事項を審議する。

第 V 章 学 会

第 15 条 本会は、毎年 1 回年次総会を開催する。会期はおうむね 10 月とするが、時宜によつて変更される。開催地は幹事会の決定による。

第 16 条 学会総会の運営に関する細目は、其のつと会長が定める。会長は幹事会にはかり、総会以外の学会を開催することができる。

第 VI 章 機関誌の発行

第 17 条 本会は別に定める機関誌規定によつて機関誌を発行する。

第 VII 章 会 計

第 18 条 本会の経費は、会費及び寄附金をもつて処理する。会計年度は、毎年 1 月 1 日に始まり、12 月 31 日に終る。前年度収支決算は評議員会の承認を得て、毎年学会総会に於て報告する。

第 19 条 名誉会員は、会費を免除される。

第 20 条 会費の年額は評議員会で決定する。

附 則

本会則は、昭和 32 年 6 月 16 日から実施する。

(昭和 35 年 12 月 18 日改正)



日 本 癌 学 会 会 費

昭和 36 年度は会費 1200 円です。会員には日本癌学会総会記事と「GANN」第 52 巻（第 1 号—第 4 号）をお送りします。

送金先は

東京都豊島区西巣鴨 2 / 2615

財団法人 癌研究会癌研究所内 日本癌学会事務所

日本癌学会の振替口座は東京 174423 番です。

日本癌学会記事・第 19 回総会

昭和 36 年 3 月 25 日 印刷

昭和 36 年 3 月 31 日 発行

編集兼発行
代 表 者

田 崎 勇 三

東京都豊島区西巣鴨 2 丁目

印 刷 者

加 藤 保 幸

東京都千代田区神田三崎町 2 / 12

印 刷 所

株式
会社 加 藤 文 明 社

東京都千代田区神田三崎町 2 / 12

発 行 所 日 本 癌 学 会

東京都豊島区西巣鴨 2 丁目 電話 東京(971) 5186~8 番

取 扱 店

東京都千代田区神田駿河台 3 / 9

電話 東京 (291) 7121~7 番 共立出版株式会社

#807

